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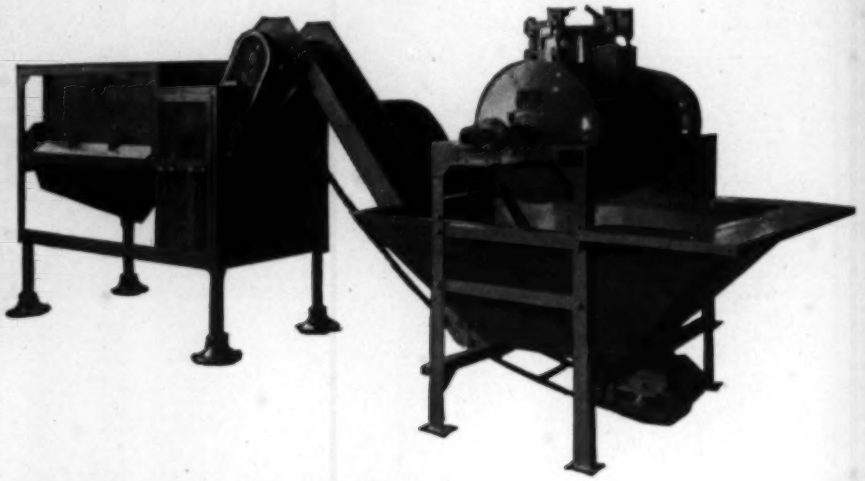

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Human Relations and the Food Technologist

Those whose education has not been so exclusively scientific as to crowd out the classics, will recall that one of the most ancient engineering projects was the attempted construction of a tower by means of which heaven was to be reached through the mere labor of climbing. It will also be remembered that the project was abandoned when the builders were struck with a "confusion of tongues." Since no modern industrial organization is a one-man show, any project requiring the integration of various sciences, arts and skills, may easily become a Tower of Babel if each participant can speak only the jargon of his own profession. The resulting failure is basically a failure in human relations for without understanding there can be no intelligent cooperation, and without cooperation there can be no success either for the individual or the corporation.

The time and effort required to orient a college graduate in industry represents wholly unnecessary waste. Colleges fail, not so much in the over-emphasis of the importance of the profession being taught, as in the under-emphasis of the importance of collateral subject matter. The resulting end product of our educational system is narrow from over-specialization, and almost completely lacking in understanding of the need for team play. And the reasons are not difficult to find.

The accumulation of human knowledge during the past generation has been almost geometric, particularly in the sciences, use of which we make constantly in the solution of business problems. Yet the sacred cow of the curriculum, the four year college course, still dominates our thinking. If thirty-six months is hopelessly inadequate to do justice to our newer knowledge of the sciences, obviously those subjects of supreme importance to the citizen, the taxpayer, and the gentleman, will be ignored.

But our concern is in connection with an attitude of mind, or perhaps more accurately, the lack of it. Education from the cradle to the grave is an individual matter. From the three R's to the doctoral dissertation it is the individual struggling to understand, absorb, and finally make his own through the medium of the brain. Obviously, this long arduous process of self-improvement, largely through individual initiative, develops a lone-wolf personality. Unless we inaugurate an exchange arrangement whereby the college instructor spends some time in industry, and the research director does some teaching, our graduates will continue to enter the business world wholly unaware of the need for substituting their feeling of self-sufficiency for one of cooperative team work. And the human mind, being what it is, will not easily reject what it learned in the process from the age of six to that of twenty-six. We must teach team play, not merely mention it.

The operation of a modern food processing plant is an amazingly complex affair. Success of the enterprize, and every person participating in it, depends upon the effectiveness with which available talent pools its brains for a united frontal attack. This is basic. Lip service will not do; it must be a conviction. Too long has the scientist remained in his Ivory Tower to the detriment of himself and of the industry he serves.

Failures in human relations are merely failures to view one's job in the proper perspective and relationship to the functions of all his associates—and governing himself accordingly. Those practicing food technology, newest of the professions, are in a favorable position to avoid the mental isolation stemming from training in only one science, for food technology is the distillate resulting from a mixture of chemistry, bacteriology, biology and engineering. But he is not immune from the tendency to play a lone hand.

If loss of time, development of friction and injured pride, were of no moment, we might let industry, a great leveler and democratizer, take its course. But we can make education the shortest distance between two points by teaching the *philosophy* of industrial research during the senior year. Only then will the young food technologist try to fit *into* the industrial scheme of things, rather than attempt to superimpose himself *on* it.

ELLERY H. HARVEY
President, I. F. T., 1946-'47

NUTRITIVE VALUE OF CANNED FOODS

In 1942 a large-scale survey of the nutritive value of canned foods was begun under the joint sponsorship of the National Canners Association and the Can Manufacturers Institute. At the end of the first year progress reports were issued from the laboratories which were cooperating in the survey.

Since that time approximately two dozen papers have appeared, dealing with the contents of several "B" vitamins, fat soluble vitamins, and minerals in canned foods, the reproducibility of assay values, and the effect of processing, storage, etc., on nutritive value.

In general it may be said that canned foods retain a large part of the original nutritive value of the foods. The distribution of nutrients between solid and liquid portions of canned foods varies widely. Both solid and liquid portion have good nutritive values.

The over-all program of the National Canners Association and the Can Manufacturers Insttiute, which has been carried through by the various cooperating laboratories, has resulted in a better understanding of the contributions made by canned foods to national nutrition, and it may be said that more is known about the nutritive values of canned foods than about any other type of processed food.—Current Research in the Science of Nutrition.

The Nutritive Value of Canned Foods. II. Changes in Ascorbic Acid of Vegetables During Storage Prior to Canning*

PAULINE PAUL, BETTY EINBECKER, LOUISE KELLEY, MARGUERITE JACKSON, LOIS JACKSON, R. E. MARSHALL, W. F. ROBERTSON, AND MARGARET A. OHLSON

Departments of Foods and Nutrition, and Horticulture, Michigan State College, East Lansing, Michigan

The authors report ascorbic acid values for corn, peas, green beans, lima beans and spinach in the raw and canned state as influenced by varying periods and conditions of storing and washing the freshly picked vegetables.

Factors affecting the nutritive value of canned vegetables are being widely studied. The aspects of the problem investigated in this study were the changes in ascorbic acid, thiamine, riboflavin and carotene in vegetables stored under varying conditions and for different times between harvesting and canning. The conditions and times of storage were representative of those used by commercial canners or considered to have some possibility of use during periods of peak load. The ascorbic acid values are reported in this paper.

A number of studies have been made on the changes in ascorbic acid content of vegetables during storage. Mack and Tressler *(9)* found that peas in the pod lost very little ascorbic acid when held at 1 and 9° C., but that there was considerable loss at 18 and 22° C. Tressler, Mack and Jenkins *(12)* reported that refrigeration retarded the loss of ascorbic acid in lima beans and that shelled lima beans lost ascorbic acid about twice as rapidly as did the unshelled ones. Eheart, et al *(3)*, concluded that lima beans should be stored in the refrigerator in the pod for maximum retention of ascorbic acid.

Dunker, Fellers, and Fitzgerald *(2)* found that corn stored in the husk either at room or refrigerator temperatures lost almost no ascorbic acid the first day and showed only a 20% loss after three days' storage. Lamb *(6)* found that green beans lost ascorbic acid rapidly during the first 24 hours at room temperature, but that subsequent losses were small. Mack and Tapley *(8)* stated that snap beans lost ascorbic acid rapidly at all temperatures, but the rate of loss was somewhat less at lower temperatures. Gordon, Griswold and Porter *(4)* found that the use of snow ice in holding green beans improved the ascorbic acid retention materially over that of beans held at room temperature. The work of Brasher, et al *(1)* indicated that for shelled lima beans, ice refrigeration was preferable to cracked ice storage. They suggested that the increased loss of ascorbic acid of the beans stored in cracked ice might be attributed to the effect of extra moisture in favoring enzyme activity. The use of a propionate solution as a dip or spray to prevent microbial growth in stored produce has been suggested by Wolford and Anderson *(13)*.

*Journal Article No. 868 (N.S.) from the Michigan Agricultural Experiment Station. This is No. XXXII in the series on the Nutritive Value of Canned Foods, supported in part by funds provided by the National Canners' Association and the Can Manufacturers' Institute.

Jackson *(5)* investigated the changes in reduced, dehydro- and total ascorbic acid during refrigerated storage of peas and spinach. She reported that peas retained nearly all their ascorbic acid for 48 hours, while spinach lost 7% of the reduced and 23% of the total ascorbic acid. Most of the samples showed a decrease in ascorbic acid during the first hour of storage, followed by an increase after three hours, then a gradual drop to or below the original value with increased time of storage.

Materials and Methods

Details concerning the growing, harvesting and handling of the vegetable crops are given in the first paper of this series, (Marshall and Robertson *10*).

Field samples were obtained by random hand picking of the field just before the crops were harvested, and were analyzed as rapidly as possible, usually within 30 minutes after leaving the field. Therefore, these samples were analyzed one to three hours before the samples marked as "prompt handling" in the storage series, since all the storage series samples went through the usual commercial handling procedures. The field samples were not canned since it was not possible to provide a large enough sample to process with cannery equipment.

Five to 10 pounds of each sample of raw vegetables were brought to the laboratory for analysis. Samples which contained dirt were washed in tap water, then dried with towels. The samples which had been washed as part of the storage treatment did not require further washing. Four ascorbic acid determinations were made on each sample of corn, green beans and lima beans, and five on each sample of peas and spinach. Total solids determinations were done in duplicate.

In the lots that were canned, one number 10 can from each storage sample was opened and drained five minutes in a large colander. Weights of the solid and liquid portions were recorded. Solids and liquids were analyzed for ascorbic acid and total solids. The ascorbic acid values for the total can contents were calculated from the ascorbic acid content of the solids and liquid, and the original weight of each contained in the can.

Because of technical difficulties, it was necessary to do total, rather than reduced ascorbic acid on the corn. The method used was that of Roe and Oesterling *(11)*. Ascorbic acid determinations for the peas, string beans, lima beans and spinach were made by the Loeffler and Ponting method *(7)*. Five percent metaphosphoric acid was used for the original blend and the blend diluted to conform with the method as described.

Total solids were determined by drying weighed samples for 48 hours in a 70° C. oven with forced air circulation.

The times indicated in the discussion of results and in

the tables correspond to those given by Marshall and Robertson *(10)* as elapsed time from harvest to processing.

Results

The ascorbic acid contents of the vegetables are listed in Tables 1 to 5. Data are given for the raw and canned vegetables, on both the wet and dry basis. The values for the canned samples are based on the total can contents.

Corn: The data obtained on corn are summarized in Table 1. The two lots of raw corn followed about the same pattern, the ascorbic acid decreasing slightly with holding, then increasing for the longest holding period. The results were about the same when calculated on the dry basis, indicating that the increase was not due to water loss. The two lots differed in maturity as well as variety, which would account for the higher level of ascorbic acid in the second lot.

The canned samples all had about the same amount of ascorbic acid remaining in them, with the exception of the "prompt handling" sample of Seneca Golden, which had the lowest ascorbic acid content in both the raw and canned samples.

TABLE 1

Ascorbic acid values for corn

Variety	Method of holding	Time hrs.	Raw		Canned	
			mg/100gm. raw weight	mg/gm. solids	mg/100gm. total can contents	mg/gm. solids
Seneca Golden	Field sample	....	9.7	....†	....*	....†
	Prompt handling	2.3	7.7	0.23	2.8	....†
	Held on wagon	5.9	8.5	0.27	4.1	....†
		8.7	8.1	0.25	4.4	....†
		25.5	9.5	0.29	4.3	....†
Golden Bantam	Field sample	....	13.6	0.48	....*	....*
	Prompt handling	2.0	13.8	0.55	4.8	0.32
	Held on wagon	4.9	12.3	0.47	4.7	0.30
		7.4	13.6	0.52	4.7	0.31
		23.8	15.3	0.56	4.4	0.30

*Not canned. †Not determined.

TABLE 2

Ascorbic acid values for peas

Variety	Method of holding	Time, hrs.	Raw		Canned	
			mg/100gm. raw weight	mg/gm solids	mg/100gm. total can contents	mg/gm solids
Thomas Laxton	Field sample	....	22.2	1.10	....*	....*
	Prompt handling	2.3	20.5	1.04	9.3	0.64
	Shade	5.5	14.1	0.94	9.8	0.76
	Washed, shade	5.5	15.1	0.82	6.8	0.48
	Unvined	4.3	19.8	1.11	....*	....*
	Running water	9.3	12.9	0.84	7.3	0.52
	Cold storage	9.4	10.8	0.57	8.6	0.60
		22.8	9.8	0.49	7.5	0.51
		46.8	6.7	0.34	7.6	0.52
	Washed, cold storage	9.4	10.4	0.64	8.3	0.58
		22.8	11.0	0.63	6.1	0.46
		46.8	10.2	0.54	5.9	0.46
	Snow ice	22.8	11.8	0.64	7.8	0.58
		46.8	10.6	0.58	4.7	0.37
Pride	Field sample	....	22.4	1.11	....*	....*
	Prompt handling	1.3	18.5	0.97	8.0	0.49
	Shade	4.8	15.0	0.70	8.4	0.49
		10.3	15.9	0.81	7.0	0.45
	Washed, shade	4.8	16.6	0.80	8.5	0.68
		10.3	16.1	0.89	5.9	0.39
	Running water	10.3	14.6	0.90	5.9	0.40
	Cold storage	10.3	17.8	0.94	8.7	0.44
		26.0	13.3	0.63	6.9	0.48
		48.3	16.1	0.79	6.2	0.39
	Washed, cold storage	10.3	15.0	....†	5.7	0.42
		26.0	12.4	0.65	11.0	0.73
		48.3	15.9	0.81	6.7	0.44
	Snow ice	26.0	13.0	0.66	5.9	0.41
		48.3	16.2	0.82	6.1	0.43
Shasta	Field sample	....	24.6	1.27	....*	....*
	Prompt handling	1.2	16.2	0.80	10.6	0.56
	Shade	4.4	16.0	0.75	10.3	0.71
		7.9	18.3	....†	10.6	0.68
	Washed, shade	4.4	15.3	0.74	11.3	0.84
		7.9	21.9	1.24	....†	....†
	Unvined	17.3	15.9	0.75	11.2	0.73
	Running water	7.9	16.2	0.95	10.9	0.74
	Cold storage	7.9	23.2	1.21	11.0	0.72
		24.2	17.5	0.83	....†	....†
	Washed, cold storage	7.9	24.0	1.35	9.7	0.63
		24.2	21.5	1.17	8.5	0.55
	Snow ice	24.2	19.5	1.00	8.9	0.58

*Not canned. †Not determined.

Peas: The ascorbic acid values for the peas are given in Table 2. The highest ascorbic acid retention in the raw Thomas Laxton peas was found in the "prompt handling" sample. The sample held 4.3 hours before vining was the next highest. Washing the shelled peas before holding increased the ascorbic acid retention.

The "prompt handling" sample of raw Pride peas was also highest in ascorbic acid retention, the second highest being the sample held 10.3 hours in cold storage. The washed samples of raw Pride peas were slightly higher than the unwashed when held in the shade, but not when held in cold storage.

The samples of raw Shasta peas increased in ascorbic acid content with increased time of holding in the shade, also when held in cold storage 7.9 hours. The highest ascorbic acid content was found in the washed sample held 7.9 hours in cold storage, and the second highest was in the unwashed sample held the same length of time.

The highest ascorbic acid values for the canned peas were: Thomas Laxton, 5.5 hours in shade; Pride, 26 hours in cold storage; Shasta, washed and 4.4 hours in shade. In most instances, the ascorbic acid of the canned samples decreased as the time of holding the raw product under any one set of conditions increased.

Green beans: Table 3 summarizes the values obtained for green beans. The ascorbic acid content of the raw green beans remained high for 5-6 hours in the shade, and 48 hours in cold storage, then decreased. The sample held 11 hours in the shade had decreased in moisture content so that the ascorbic acid content was lowered when calculated on the dry basis. The use of snow ice on the beans in cold storage did not increase the retention of ascorbic acid.

TABLE 3

Ascorbic acid values for green beans (Landreth's Stringless Greenpod)

Method of holding	Time, hrs.	Raw		Canned	
		mg/100gm. raw weight	mg/gm solids	mg/100gm. total can contents	mg/gm solids
Field sample	....	29.2	2.71	....*	....*
Prompt handling	1.6-2.1	26.1	2.51	7.9	0.94
Shade	5-6	26.0	2.44	8.4	0.81
	11	26.3	2.10	7.2	0.77
	23-24	22.0	1.89	6.6	0.74
	48	18.1	1.57	5.1	0.55
	11	27.4	2.66	8.8	0.97
	23-24	26.7	2.29	8.8	0.98
Cold storage	48	28.4	2.69	9.5	0.84
	72	25.0	2.18	7.5	0.82
	120	19.9	1.86	6.7	0.68
	48	27.7	2.67	8.3	0.91
Snow ice	72	22.0	2.29	8.3	0.85
	120	19.1	1.88	7.2	0.85

*Not canned.

The values for the canned beans showed about the same trend as for the raw beans, the ascorbic acid content holding up for the samples stored 5-6 hours in the shade and 48 hours in cold storage, then decreasing. The use of snow ice apparently improved the retention of ascorbic acid in the canned samples which had been held the longest period (120 hours) in cold storage prior to canning.

Lima beans: The ascorbic acid content of the raw lima beans decreased gradually with increased holding time under any conditions (Table 4). Washing the beans after vining improved the retention of ascorbic acid. The use of a 10% Calcium propionate dip decreased the retention of ascorbic acid. Holding in running water or in snow ice was not beneficial. The unwashed beans could be held 8.2-9.2 hours in the shade or 48 hours in cold storage with only small loss of ascorbic acid as compared to the "prompt handling" sample. The washed beans held up well for 24 hours in the shade.

TABLE 4

Ascorbic acid values for lima beans (Thorogreen)

Method of holding	Time hrs.	Raw		Canned	
		mg/100gm. raw weight	mg/gm solids	mg/100gm. total can contents	mg/gm solids
Field sample	....	41.9	3.43	....*	....*
Prompt handling	1.5-1.9	32.7	1.83	13.3	0.45
Shade	5.2	30.8	1.62	12.4	0.46
	8.2-9.2	31.8	1.52	11.4	0.40
	24	26.9	1.36	10.2	0.34
Running water	8.2-9.2	26.2	1.51	9.1	0.33
	24	22.5	1.29	8.9	0.32
Washed, shade	8.2-9.2	33.3	1.99	10.5	0.38
	24	30.7	1.76	10.6	0.39
10% Ca Pr†, shade	24	23.8	1.22	10.8	0.39
Cold storage	24	28.6	1.48	11.2	0.40
	48	28.7	1.42	....*	....*
	120	24.4	1.26	....*	....*
Washed, cold storage	120	26.1	1.44	....*	....*
10% Ca Pr†, cold storage	120	20.6	1.06	....*	....*
Snow ice	24	24.0	1.25	10.7	0.38
	120	18.9	0.90	....*	....*

*Not canned. †Calcium propionate.

The ascorbic acid values for the canned lima beans were more nearly alike than were the values for the raw beans, but the trends were the same.

Spinach: The two runs of spinach differed in original ascorbic acid content, due to differences in variety, planting date, and soil. However, the pattern of change in ascorbic acid content with holding was the same, all the samples retaining a high level of ascorbic acid except those held longer than 24 hours in the shade. There was little difference between holding in baskets or in piles in the shade. These results are summarized in Table 5.

Most of the canned samples were lost through spoilage, so no ascorbic acid values are reported for these.

TABLE 5

Ascorbic acid values for spinach

Method of holding	Time hrs.	Raw			
		Upland		Muck	
		mg/100gm. raw weight	mg/gm solids	mg/100gm. raw weight	mg/gm solids
Field sample	....			31.8	4.90
Prompt handling	2			31.4	4.16
Overnight in field	17	51.0	6.69		
Shade, baskets	7			32.2	4.75
	21	54.5	6.80		
	24			32.3	4.62
	37	44.3	6.40		
	72			18.4	2.55
Shade, pile	24			32.6	4.82
	37	47.9	6.19		
Cold storage	24			34.7	5.13
	37	57.7	7.13		
	61	56.0	7.32		
	72			31.0	5.22
	133	56.8	7.91		

Summary

The changes in ascorbic acid content have been determined for vegetables held different lengths of time under various conditions. The data were calculated on both the wet and the dry basis.

Under the conditions of this series of experiments, the following conclusions seem tenable:

1. Where the length of storage was comparable, cold storage usually gave better ascorbic acid retention than storage at outdoor temperatures, especially as the holding time increased.

2. Holding peas and lima beans in running water generally decreased the ascorbic acid retention.

3. Washing before holding definitely increased the ascorbic acid retention of the lima beans, but made little difference in the peas.

4. The use of a 10% Calcium propionate wash decreased the ascorbic acid retention in the lima beans, the only vegetable on which this was tried.

5. Ascorbic acid retention in corn was not much affected by any of the treatments used.

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Apparatus for the Measurement of the Rate of Heat Penetration in Canned Foods

O. F. ECKLUND

General Research Laboratory, American Can Company, Maywood, Illinois

A description of specialized thermocouple equipment used for heat penetration measurements is given, as well as a brief history of the development of heat penetration equipment. The equipment, now commercially available, is designed to permit normal filling and closing of the test containers on commercial equipment.

Although the mathematical analysis of thermal processes is well covered in scientific literature, little information can be found regarding the equipment needed to make the heat penetration determinations. Highly specialized equipment has been developed for this purpose and is now commercially available.[a]

Early heat penetration measurements were accomplished by maximum reading thermometers inserted into cans. This method, of course, revealed only the maximum temperature attained at the centers of the cans and gave little indication of the time-temperature relationships required for process calculation. In some instances thermometers were inserted into cans through packing glands so that the can center temperatures could be read periodically during the heating and cooling cycles if the processing was done in an open bath. Some tests were made in which a second packing gland sealed the thermometer through the side of a retort so that the temperature could be observed during a pressure process.

In recent years, the thermocouple has replaced the thermometer for use in determining the rate of heating and cooling of canned foods. Using this device the internal can temperature may be read at any time on a potentiometer which is connected to the thermocouple by a lead wire.

At first the thermocouples were made with the lead

[a] The author can supply information relative to the availability of equipment described in this paper.

wires attached and the thermocouples were inserted into the cans through stuffing boxes. This arrangement greatly complicated the filling and closing of the test cans. It was not possible to close the test cans with the thermocouples attached on high speed closing equipment. A great advance in convenience was made some years ago when the connector type thermocouple was developed. As the name implies, the lead wire was detachable from the thermocouple. This development permitted joining several lead wires into a cable and sealing the cable through the retort body with a packing gland. Since the thermocouple projected from the test can, it was not possible to close a test can with this thermocouple attached on high speed closing equipment.

The newest type of heat penetration thermocouple, in addition to having the connector feature, is so designed that the thermocouple and the receptacle which holds it do not project from the side of the can. (Fig. 1). This

FIG. 1. Thermocouple installed in can. Design of thermocouple permits normal closure of can with high speed commercial closing equipment.

permits normal closure of the can on commercial closing equipment. The thermocouples are made in various lengths to fit various can diameters, or a long thermocouple, by use of a packing gland, may be used as an adjustable length thermocouple. However, when long thermocouples with packing glands are used, special care in filling and closing the container is necessary.

Fig. 2 illustrates the complete equipment required for a heat penetration determination. The essential units are:

1. Thermocouples and receptacles for holding the thermocouples in the cans.

FIG. 2. Cans located in retort ready for heat penetration test. In this case a seven wire cable is used (6 cans and one retort thermocouple). Note retort stuffing box, selector switch and portable potentiometer.

2. Heat penetration cable consisting of lead wires fitted at one end with connectors for attaching to thermocouples, the other ends being attached to a selector switch. A packing gland is located in the middle of the cable to seal the wires through the retort wall.

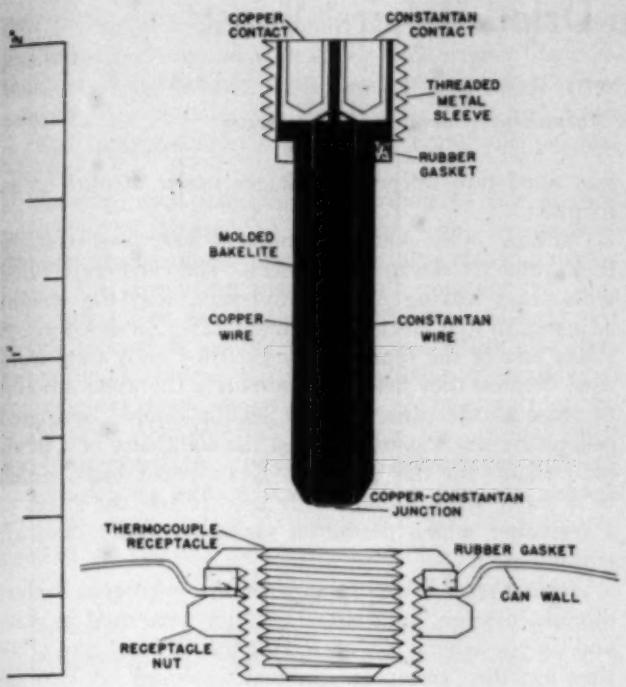
3. A potentiometer for indicating the temperatures obtained.

The thermocouples, shown in cross section in Fig. 3, are constructed of molded thermosetting plastic containing fine (30 B & S gauge) copper and constantan wires. The wires are soldered at the hot junction end and attached to drilled copper and constantan inserts at the threaded end of the thermocouple. The tubular section of the thermocouple is $\frac{1}{4}$" in diameter. The threaded section is $\frac{5}{16}$" long and is threaded $\frac{7}{16}$" x 24 threads per inch. The thermocouple is sealed to the receptacle and the receptacle to the can by means of rubber gaskets.

Tests were made to find whether this type of thermocouple would give erroneous readings because of heat conducted into the can by the thermocouple wires or by the receptacle. It was found that no significant error resulted except for small cans containing conduction heating products. One minute added to the slope, f_h, after Ball *(1)*, of the heating curve for conduction heating products in $2^{11}\frac{1}{16}$" diameter and smaller cans corrects for this error.

The heat penetration cable is usually made up of from five to ten lead wires about 12 feet long. Enameled and insulated 20 gauge wires are usually used as lead wires, heavier wires being too stiff and lighter wires being too fragile. Because a large temperature differential exists between the two ends of the lead wires, it is important that the lead wire be of high accuracy, or at least that the wires in the cable be matched. Asbestos insulation is preferred to fibre glass insulation for this application because it is more resistant to abrasion.

Connector pins are attached to each lead wire as shown in Fig. 4. The pins, made of hard drawn copper and constantan wire, are drilled on one end to admit the lead wire and are slotted on the other to form a spring contact when inserted in the thermocouple. They



THERMOCOUPLE FOR MEASURING HEAT PENETRATION IN CANNED FOODS

FIG. 3. Cross section of thermocouple and receptacle used for measuring heat penetration in canned foods.

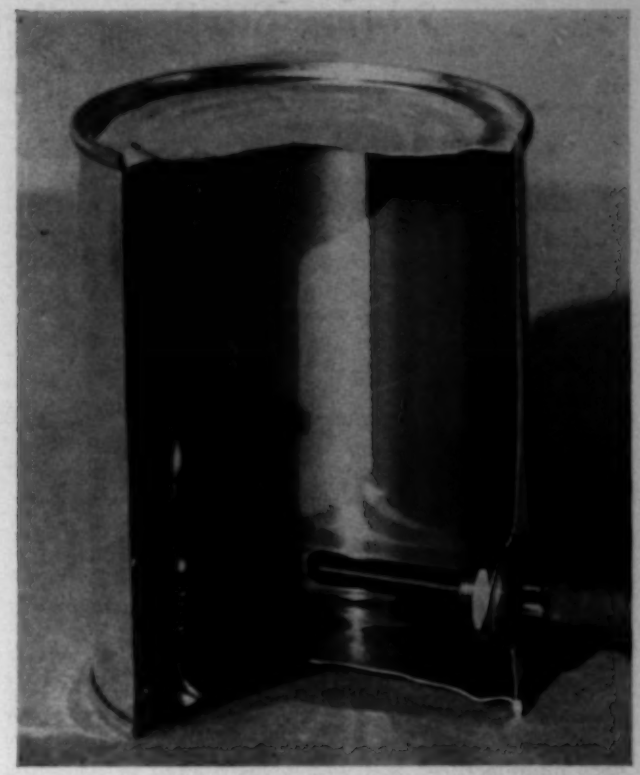


FIG. 4. Lead wire with connector attached plugged into thermocouple. Installation of thermocouple near bottom of can is proper for convection heating products.

are held firmly on the wires by squeezing the pins at two points rather than by soldering. The pins may be wrapped with string, coated with bakelite cement and baked as shown or a connector assembly may be used.

The selector switch to which the lead wires connect is of the two-pole multi-point variety. The switch need not be of the compensating type (constructed of the thermocouple metals) provided it is enclosed in a container to prevent differences in temperature at the lugs where the lead wires are soldered. Small radio switches enclosed in 2 x 4 x 4 inch metal boxes have been found to be entirely satisfactory. A single length of lead wire connects from the switch to the potentiometer.

A potentiometer (Wheatstone bridge) type of instrument is required to indicate the temperatures existing at the thermocouple hot junctions. This type of instrument indicates the thermocouple output voltage independent of the resistance of the lead wire. Variations in resistance of the lead wire introduce errors if a millivoltmeter type instrument is used.

The voltage developed by a thermocouple is not an absolute value depending on the temperature of the hot junction alone. Instead, the voltage developed depends on the difference in temperature between the hot junction and the cold junction (the point where the lead wires connect to wires of one metal). This cold or reference junction may be placed in an ice bath or may be located at room temperature. If the reference junction is kept in an ice bath, the voltage developed by the hot junction is almost in proportion to the number of degrees the hot junction is above freezing. Potentiometers reading in millivolts can be used with the cold junction in an ice bath or potentiometers may be obtained to read the hot junction temperature directly when using an "ice bottle" reference junction. Potentiometers calibrated for use without an "ice bottle" have either automatic or manual cold junction compensation. These instruments are available calibrated directly in degrees F. Of the above instruments, the "ice bottle" type is least expensive and probably most accurate. The automatic cold junction type, however, if of sufficient accuracy and is most convenient.

Summary

Special thermocouples and associated equipment have been developed for measuring the internal temperatures of containers during heat treatment. The equipment now available was designed to permit filling and closing of the containers in the normal manner on commercial equipment.

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Storage Studies on Active Dried Bakers' Yeast[a]

ROY E. MORSE[b] AND CARL R. FELLERS

Department of Food Technology, University of Massachusetts, Amherst, Massachusetts

Active dried yeast first appeared in quantity during World War II; 6,000 tons were produced in 1945. Storage conditions for this yeast were not widely known. Storage tests in sealed containers containing air, oxygen, nitrogen, CO_2, and partial vacuum, stored at various temperatures ranging from —20°C. to 37°C. were carried out. The reaction of yeast exposed to sunlight was also tried. Results showed that temperature had the greatest influence on shelf-life of active dried yeast. Light, packaging material and atmosphere appeared to have little effect. Oyaas, Johnson, Peterson and Irvin found no difference among samples stored in CO_2, nitrogen, or in vacuum, but deterioration was most rapid in air and was dependent on the amount of air and method of manufacturing. Preliminary investigations by the authors point strongly to the importance of the humidity of the packaging atmosphere.

Dried yeast has been used for many years; the Near East countries have used dried yeast which they procured from Germany. Active dried bakers' yeast has been manufactured and widely distributed in Australia for many years. One of the best known brands, "DRIBARM" is manufactured by Mauri Bros. & Thompson, Sydney, N.S.W. This yeast was packed with malt flour and ammonium salts to aid in rehydration and activation. In 1944 alone the U. S. Army procured 1,611,000 lbs. of dry yeast from Australian producers. Its development in the United States was greatly accelerated by World War II. Boruff and Van Lanen *(3)* have shown its tremendous growth to be from 900 tons in 1942, to 6000 tons in 1945, the peak year. Because of this rapid development its use outstripped published technical knowledge. Thiessen *(7)*, and an anonymous publication by the Corps of Quartermaster *(1)* have stated that low storage temperatures are desirable for active dried yeast. A recent publication by Oyaas, Johnson, Peterson, and Irvin *(6)* studied the storage of active dried yeast produced by four different manufacturers and they concluded that storage in the absence of air was best, and that storage in an excess of air was most destructive to viability. Atkin, Kirby, and Frey *(2)* have patented the storage of active dried yeast in an atmosphere of carbon monoxide, claiming improved storage life. Since further information was needed about correct storage conditions for this new product it was decided to investigate the factors which influence its viability during storage.

Experimental

A one hundred pound batch of dried yeast was obtained and immediately upon receipt was filled into No. 10 tin cans, which were sealed, and stored at 5° C. until needed. For the following storage study the yeast was filled into different packages under conditions as follows:

Package: One-pound-flat tin cans were used to study their value as storage containers. The cans were filled with yeast leaving ¼ in. headspace, and the weight adjusted so that all cans contained 225 g. of dried yeast. Glass jars of the type usually used for baby food were used because they held approximately the same amount of yeast as the cans. A heat sealing double laminated cellophane bag was used to test the suitability of a flexible package for this product. Kraft paper bags, sealed by a staple after folding, were used to test the effect of a container which permitted moisture, vapor, and air transmission.

Atmosphere: Atmospheres of air, nitrogen, carbon dioxide, oxygen, and partial vacuum were used in glass and tin containers. To obtain the gas atmospheres other than air, the "sweeping" method was used. A vacuum was drawn, then broken by the desired gas three times; after the last gas filling the container was sealed. A vacuum of about 18 in. was obtained with the equipment available.

Light: To study the effects of light, duplicate jars were packed and sealed. One jar was then placed in a No. 2 can which had five large holes punched through its bottom and the can was sealed. The canned and uncanned jars were placed side by side upon a laboratory shelf about five feet from a window with southern exposure.

Temperature: The temperatures used during this study were, —20°, 5°, 25°, and 37° C. For the lowest temperature a home style freezing cabinet was used; a refrigerator was used for the 5° studies; a laboratory cabinet was used for the 25° studies; and an incubator for the 37° C. studies.

Time: Times for examination were selected on the basis of estimated maximum change in viability, the samples stored at higher temperatures were examined more frequently than those at lower temperatures. A summary of the storage study pack is shown in Table 1.

TABLE 1

Summary of Active Dried Yeast Storage Study

Stored at —20° and 5° C. for 1, 2, 4, and 6.5 Mos.			
Container: Can			Kraft Paper Bag
Atmosphere: Air, CO_2, N and Vacuum			Air
Stored at 25° C. for 1, 2, 4, 8, 16, and 20 Weeks			
Container: Glass Jar		Laminated Cello Bag	Kraft Paper Bag
Atmosphere: Vacuum, N, Air		Air	Air
Light and Dark			
Stored at 37° C. for 7, 14, 21, 25, 28, 32, 36, and 48 Days			
Container: Can			
Atmosphere: Air, Vacuum, CO_2, and O_2			

Methods

At the conclusion of the storage period all containers were allowed to come slowly to room temperature. Those stored at the colder temperatures, —20°, and

[a] Contribution No. 696, Massachusetts Agricultural Experiment Station.

[b] Present address, Monsanto Chemical Co., Anniston, Ala.

5° C., usually required about 24 hours. Cans were examined for vacuum with a dial type gage. The contents of the container were thoroughly mixed before sampling by pouring the dried yeast back and forth several times between a large beaker and the original container.

Viability was determined according to the method described by Morse *(5)* which employs a yeast-flour mixture sealed into a pipe which is attached to a manometer, the pipe being immersed in a constant temperature bath. The pressure generated over a 2-hour test period is used as an index of viability. With all determinations, a control was run, consisting of yeast from the original batch stored at 5° C. At the start of the study several samples of yeast from the original batch were examined for viability, and subsequent viability determinations were referred to this figure. All storage analyses were run in duplicate and the average was used as the viability number.

Results

The effect of storing active dried yeast under different conditions at —20° and 5° C. is shown in Table 2.

TABLE 2

Viability Index of Active Dried Yeast Stored at —20° C.

Sample Description	Storage Time in Months			
	1	2	4	6.5
Can, vacuum	635	597	651	795
Can, carbon dioxide	498	665	604	764
Can, nitrogen	480	509	622	812
Can, air	573	688	634	709
Kraft paper bag	480	645	615	767
Control	584	650	599	654

Viability Index of Active Dried Yeast Stored at 5° C.

Sample Description	Storage Time in Months			
	1	2	4	6.5
Can, vacuum	526	552	673	655
Can, carbon dioxide	571	656	624	636
Can, nitrogen	678	507	624	727
Can, air	615	613	664	644
Kraft paper bag	566	588	656	571
Control	669	606	683	654

Viability index at start of test period—640.

Examination of Table 2 shows that all samples stored at —20° had better viability after 6.5 months than the control which had been stored at 5° C. in an atmosphere of air. It was considered that there was no significant difference between all samples stored at —20° C.

Examination of the data shows that no significant difference exists between all samples stored at 5° C. and the control. The air atmosphere stored samples agreed quite closely with the control analyses which serves as a good check since they were in reality duplicates. The sample stored in a kraft paper bag was slightly below the control in viability, but not enough to be significant.

The results of the analyses conducted upon yeast at 25° C. are shown in Table 3.

Examination of these data shows that packaging atmosphere appears to have little effect upon the viability of the stored yeast. However, all samples showed 10-20 percent lower viability than the control, and since all samples stored at —20° C. showed approximately 10-20 percent higher viability it becomes apparent that lower storage temperatures have a favorable effect upon viability retention.

TABLE 3

Viability Index of Active Dried Yeast Stored at 25° C.

Sample Description	Storage Time in Weeks					
	1	2	4	8	12	20
Glass, vacuum, dark	613	565	259	581	507	639
Glass, vacuum, light	619	616	434	616	623	610
Glass, nitrogen, dark	600	523	539	622	555	588
Glass, nitrogen, light	588	592	498	532	540	514
Glass, air, light	605	449	389	489	527	589
Kraft paper bag	630	527	607	506	565	550
Laminated cellophane bag	594	680	552	458	495	262
Control	629	631	683	529	599	677

The samples stored at 37° C. showed viability numbers as shown in Table 4.

TABLE 4

Viability Index of Active Dried Yeast Stored at 37° C.

Sample Description	Storage Time in Days							
	7	14	21	25	28	33	37	50
Vacuum	....	262	455	180	310	62	12	11
CO_2	....	446	199	126	96	370	185	13
Air	599	394	243	225	95	408	305	65
Oxygen	507	331	225	....	....	....	....	....
Control	575	521	509	498	601	702	617	619

The analyses show that while the higher storage temperatures were more destructive of viability than the lower temperatures, the packaging atmosphere had little demonstrable effect. Samples stored in an atmosphere of oxygen deteriorated more rapidly than those stored in an atmosphere of air. Concurrent with the oxygen storage test a study was made of the atmosphere changes within a stored can of active dried yeast. Several cans of dried yeast were sealed in an air atmosphere and stored for 5 months at 25° C. Analysis of the can atmosphere according to Lewis *(4)* showed the composition of the atmosphere to be 12.3% carbon dioxide, 1.2% oxygen and (by difference) 86.5% nitrogen. From the above data it is apparent that active dried yeast continues to respire in storage; consuming oxygen and liberating carbon dioxide.

A preliminary study was made to investigate the effect of the humidity of air upon stored dried yeast. Different concentrations of sulphuric acid were used to produce the several relative humidities. The results of this preliminary study indicate that the higher humidities are harmful to the viability of stored dried yeast. Sample data are given in Table 5.

A comparison of the air atmosphere samples stored at different temperatures is shown in Figure 1.

TABLE 5

Effect of Humidity on Viability Index of Stored Dried Yeast

Relative Humidity	Storage Time in Days at 37° C.	
	2	6
100	528	308
97	512	387
58	504	431
3	587	464
*Control	509	498

*Control sample stored in closed container at 5° C. and 75% ave. rel. hum.

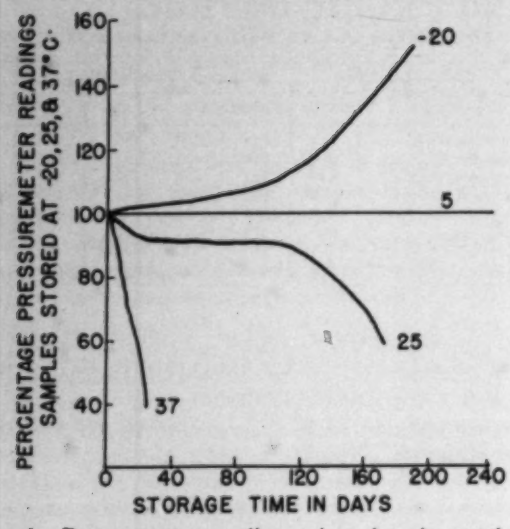

FIG. 1. Pressuremeter readings given by air-stored active dried yeast shown as percentage of similar samples stored at 5° C.

Summary

Examination of the above data shows that of the storage factors studied, temperature had the greatest influence on the viability of dried bakers' yeast. Light, packaging material, and atmosphere appeared to have little effect. This is in agreement with Oyaas, Johnson, Peterson and Irvin *(6)* who found no difference among samples stored in carbon dioxide, nitrogen, or in vacuo. However, they found that deterioration was more rapid in air, and was dependent upon the method of manufacture, and on the amount of air. Preliminary investigations point strongly to the importance of the humidity of the packaging atmosphere.

Acknowledgments

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Factors Affecting the Beading and Leakage of Soft Meringues

E. ELIZABETH HESTER AND CATHERINE J. PERSONIUS
New York State College of Home Economics at Cornell University, Ithaca, New York

A desirable degree of coagulation is suggested as an important factor in controlling the amount of leakage and beading in a soft meringue. Excessive leakage was observed under conditions which would tend to result in incomplete coagulation, viz. with meringues baked on a cold base, about 59°F. (15°C.), and with those baked for about 4½ minutes at a high temperature, 425°F. (218°C.). Excessive beading occurred under conditions which would favor overcoagulation, viz. with meringues baked on a hot base, about 131° to 140°F. (55° to 60°C.), and with those baked for ten minutes at 325°F. (163°C.) or for seven minutes at 375°F. (190.5°C.). Egg quality and the addition or omission of potassium acid tartrate was of little significance in affecting leakage. The addition of potassium acid tartrate, especially to eggs of lower grades, tended to increase the extent of beading.

Beading and leakage of soft meringues are problems for which no wholly satisfactory solution has been found. A review of the literature reveals little concerning controlled studies on soft meringues. The studies reported are primarily ones on egg quality, egg white foams, and the meringue-base of angel food cakes. The work described in this paper was undertaken in an attempt to discover the effect of several factors on the qualities of soft meringues as used for toppings of pies or puddings. The factors studied were quality of eggs, addition of potassium acid tartrate, type and temperature of the base on which a meringue is baked and baking time and temperature. The qualities observed were beading, leakage, the ease of cutting, surface appearance, and flavor. In this paper only the effects on beading and leakage will be discussed with brief mention of the ease of cutting.

Experimental Procedure

In preliminary experiments the effect of proportion of ingredients and of mixing procedure upon the specific gravity and stability of egg white foams and upon the palatability of meringues made by the foams were studied. The most desirable meringues were made by the addition of about 0.25 g. of salt to 30 g. of egg white at the beginning of the beating, and the addition of 25 g. of sugar when the white had been beaten to a relatively stiff foam with a specific gravity of 0.15 to 0.17. These proportions of salt and sugar to egg white were used throughout this study.

Four qualities of eggs were used: New York Grades Fancy, A, B, and C. The eggs of each grade were treated with three levels of potassium acid tartrate, viz.

no potassium acid tartrate, 0.25 g. and 0.5 g. to 30 g. of egg white. Each meringue mixture was divided into nine equal parts, three being placed on hot bases, approximately 131° to 140° F. (55° to 60° C.), three on cold bases, approximately 59° F. (15° C.), and three on paper. One meringue on each type of base was baked at each of the following temperatures: 325° F. (163° C.) for about 10 minutes, 375° F. (190.5° C.) for about 7 minutes, and 425° F. (218° C.) for about 4½ minutes. Each series of tests was repeated four times making a total of 432 meringues. The design of the experiment made possible statistical treatment of the data.

The eggs were obtained from the Poultry Department of Cornell University. They were gathered from one breed of hens (Leghorn) which had been fed the same food. The eggs for one series of tests were from one or no more than two days production. These eggs were stored at controlled temperatures until the desired quality was obtained as judged by candling tests.

For each series of tests a uniform sample of egg white weighing 720 g. was used. With each level of potassium acid tartrate the completed meringue mixture was a quantity sufficient for nine meringues weighing 30 g. each.

To obtain a uniform mix the white was beaten from 45 to 60 seconds, at medium speed using a rotary-type electric beater. pH determinations were made on five samples of the mixture using a Beckman glass electrode pH meter. Egg-white mix was removed for the first day's test; the remaining portion, sufficient for two additional tests, was stored in the refrigerator in a tightly sealed glass jar for use the following day. Before using the stored mix, five more pH determinations were made.

When no potassium acid tartrate was to be used, salt was added to the white at the beginning of the beating. The white was beaten at medium speed to a specific gravity of 0.15 to 0.17; beating time ranged from two minutes to three minutes and 25 seconds. The foam was relatively fine and stable with small air cells and formed rounded peaks when the beater was withdrawn. The sugar was gradually added during the following 90 seconds, and the beating continued until the meringue had a specific gravity of 0.22 to 0.24. Beating time varied from five to seven minutes. The finished meringue was stiff, but not dry, and formed rounded peaks when the beater was withdrawn.

When potassium acid tartrate was used it was added when the foam reached the frothy stage in order to obtain a more even distribution throughout the egg mixture. Five pH determinations were made, then salt was added and beating continued as described above.

To determine foam stability 10 g. of the meringue were placed in a glass funnel and covered with a watch glass. At the end of one hour the amount of liquid which had drained was recorded.

Starch-thickened water was used as a base on which to bake the meringues. The mixture was poured into aluminum pie pans, 9.5 cm. in diameter and having a capacity of 125 ml. The fillings to be used as cold bases were cooled in the refrigerator; those to be used as hot bases were set in a pan of simmering water, approximately 194° F. (90° C.). As a control, samples of each meringue were baked on heavy brown paper. Each meringue was baked to approximately the same degree of brownness, a delicate white with the irregular surfaces a golden brown. Exact baking times were recorded.

The meringues were cooled to room temperature and, from 30 minutes to two hours after baking, were judged subjectively.

Results

Snedecor's *(4)* method for determining variance between classes and within classes was used to determine the significance of the variations as judged subjectively. Tables 1 and 2 show the effect of the variables on meringue leakage and Tables 1 and 3 the effect on beading, the two points for which this study was primarily designed.

Quality of Eggs. Statistical analysis of the subjective data indicates by odds greater than 99 to 1 that the grade of eggs used affected the amount of leakage of the baked meringues. The meringues made from the low quality eggs showed the least leakage, a slight increase being noted with increase in quality. Only the differences between Grades Fancy or A and Grade C are significant at the one percent level.

The extent of beading was not directly affected by the quality of eggs. The interaction between quality of eggs and the level of potassium acid tartrate will be discussed later.

The quality of the eggs appeared to have no significant effect on the time required to beat the white to a given volume as measured by specific gravity. Meringues made from eggs of various qualities which had been beaten to approximately the same specific gravity showed little difference in foam stability.

Levels of Potassium Acid Tartrate. The leakage of the baked meringues was not significantly affected by the levels of potassium acid tartrate although its addition had a stabilizing effect on the foam, stability being slightly less with the addition of 0.5 g. of the acid substance than with 0.25 g. This is in general agreement with the findings of Barmore *(1)* who reports that the proper amount of acid will reduce the rate of collapse of an egg white foam from two to three times what it would have been in the absence of the acid.

The amount of beading tended to increase with an increase in the amount of potassium acid tartrate. The addition of 0.25 g. of the acid substance to 30 g. of egg white did not increase beading significantly (odds of 99 to 1), but the largest amount of acid substance did cause a significant difference.

The quality of eggs had no apparent effect on the amount of beading, but a strong interaction between the acid salt and grade of eggs was observed. In general, with the highest level of potassium acid tartrate, the amount of beading increased with a decrease in egg quality. This trend was not observed when the acid

TABLE 1

Variance Analyses of the Data for Beading and Leakage in Soft Meringues

Source of Variance	Leakage		Beading	
	d.f.	Mean Square	d.f.	Mean Square
Grade of eggs	3	16.0228[b]	3	0.1893
Level of KHTar	2	10.0435	2	74.7211[b]
Baking base	2	1133.6810[b]	2	516.0096[b]
Baking temperature	2	57.2810[b]	2	16.3830[a]
Interactions				
Grade: KHTar	6	6.3798	6	14.1351[b]
Grade: Baking base	6	7.5980	6	1.3032
Grade: Baking temp.	6	4.4542	6	3.3299
KHTar: Baking base	4	5.1368	4	10.5418[a]
KHTar: Baking temp.	4	5.2647	4	6.1002
Bases: Baking temp.	4	4.8882	4	6.8597
Higher interactions and replicate error	69	3.8361	69	3.7184

[a] Significant at 5 percent level (odds 19:1).
[b] Significant at 1 percent level (odds 99:1).

TABLE 2

Effect of Variation of Grade of Eggs, Level of Potassium Acid Tartrate, Baking Base and Baking Temperature on the Leakage of Soft Meringues

Variance	No. of Samples[c]	Grade Fancy Mean Score[c]	Grade A Mean Score[c]	Grade B Mean Score[c]	Grade C Mean Score[c]	Mean of Total Score[c]
No KHTar	9	11.25	12.79	12.08	11.33	11.86
0.25 g. KHTar	9	12.59	11.53	11.10	10.71	11.48
0.5 g. KHTar	9	12.42	11.14	10.73	8.98	10.82
Hot base	9	8.34	7.68	6.03	5.33	6.85
Cold base	9	18.14	18.29	18.37	15.84	17.66
Paper	9	9.78	9.49	9.52	9.84	9.66
325° F.	9	10.73	11.36	10.52	8.66	10.32
375° F.	9	12.48	11.31	10.82	9.64	11.06
425° F.	9	13.04	12.79	12.57	12.71	12.78
Mean of total score		12.08	11.82	11.31	10.34	

At the 1 percent level the least difference for significance between the baking temperatures is 1.22; between the grades of eggs, is 1.41. The difference between the levels of potassium acid tartrate is not significant.

[c] The data were analyzed in such a way that the scores of the four replicates were totaled and treated as one sample; therefore, each sample number actually represents four meringues. Since each meringue was rated by 3 judges each mean score represents 108 subjective ratings and each mean of total score 432 ratings. The meringues receiving the lowest scores showed the least leakage.

TABLE 3

Effect of Variation of Grade of Eggs, Level of Potassium Acid Tartrate, Baking Base and Baking Temperature on the Beading of Soft Meringues

Variance	No. of Samples[d]	Grade Fancy Mean Score[d]	Grade A Mean Score[d]	Grade B Mean Score[d]	Grade C Mean Score[d]	Mean of Total Score[d]
No KHTar	9	7.86	6.85	5.59	5.85	6.54
0.25 g. KHTar	9	7.25	7.16	8.77	6.85	7.51
0.5 g. KHTar	9	7.94	9.61	9.18	10.76	9.37
Hot base	9	12.60	12.00	11.91	12.18	12.17
Cold base	9	5.29	5.51	5.48	5.46	5.44
Paper	9	5.16	6.11	6.15	5.81	5.81
325° F.	9	7.74	7.26	8.24	8.72	7.99
375° F.	9	8.26	8.55	8.65	8.02	8.37
425° F.	9	7.05	7.81	6.65	6.72	7.06
Mean of total score		7.68	7.87	7.85	7.82	

At the 1 percent level the least difference for significance between levels of potassium acid tartrate and between the types of bases is 1.2. At the 5 percent level the least difference for significance between baking temperatures is 0.9. The difference between grades of eggs is not significant.

[d] The data were analyzed in such a way that the scores of the four replicates were totaled and treated as one sample; therefore, each sample number actually represents four meringues. Since each meringue was rated by 3 judges each mean score represents 108 subjective ratings and each mean of total score 432 ratings. The meringues receiving the lowest scores showed the least beading.

substance was not used nor with the 0.25-gram level of cream of tartar.

Type and temperature of base. The most salient results of the experiment were observed with the different types of bases on which the meringues were baked. In respect to leakage, a significant difference between each type of base was noted with the greatest leakage from meringues baked on a cold base.

In respect to beading a highly significant difference was found between the hot base and the other two but no difference was noted between the cold base and the paper.

Baking time and temperature. The amount of leakage was observed to increase with each increment in baking temperature, with a significant difference between the lowest and highest baking temperatures. On the other hand, the baking time and temperature did not greatly affect the amount of beading, although meringues baked at 375° F. (190.5° C.) showed the greatest amount, and at 425° F. (218° C.) the least.

The baking temperature and time was the only variable that significantly affected the stickiness of the meringues on cutting. The degree of stickiness decreased with each increase in oven temperature, and the difference between the lowest temperature of 325° F. (163° C.) and the highest, 425° F. (218° C.) was highly significant. Conversely there tended to be an increase in tenderness with an increase in baking temperature.

Discussion

The results observed suggest that factors which contribute to a uniform and desirable degree of coagulation of egg proteins tend to decrease the amount of leakage and beading in a soft meringue.

Leakage. Several of the results of this experiment support the theory that one important factor in the leakage of meringues is incomplete coagulation of the egg white proteins. The excessive leakage observed in meringues baked on a cold base may be explained by the failure to heat the undersurface of these meringues sufficiently to bring about coagulation of the protein in the time required to brown the upper surface satisfactorily. Furthermore, since the base was cold these meringues cooled quickly on removal from the oven. An attempt was made, by means of thermocouples, to record the internal temperature of each meringue immediately before removal from the oven. All readings could not be used owing to mechanical difficulties. From the readings which appeared to be reliable, the maximum internal temperature reached in any of these meringues was about 158° F. (70° C.). According to Lowe *(3)* undiluted egg white protein coagulates at about 60° to 65° C. (140-149° F.). The work of Barmore *(2)* shows that sugar elevates the coagulation temperature of egg white protein, the addition of 70 to 90 g. of sugar to 100 g. of egg white causing an increase of 27° F. (15° C.). From these data it is reasonable to assume that the temperature maintained in the meringues baked on cold bases did not bring about adequate coagulation.

The small amount of drainage observed in meringues baked on hot bases suggests that the internal temperature attained in these products, and maintained for some time after removal from the oven owing to the relatively slower cooling, assured the proper degree of coagulation. This is supported by the observation that these meringues reached an internal temperature at least 18° F. (10° C.) higher than their replicates baked on cold bases.

The amount of leakage of meringues baked on paper was less than for samples baked on cold bases but greater than for those baked on hot bases. The data indicate that the internal temperature reached in these meringues was if anything slightly higher than that of the meringues baked on hot bases. The meringues baked on paper cooled quickly, however, suggesting that the time-temperature relationship was not favorable to adequate coagulation.

The data of this study indicate that adequate coagulation of meringues may be attained within a fairly wide range of baking temperatures provided the baking time is properly adjusted. For any given base used the leakage was less pronounced in meringues baked at 325° F. (163° C.) for about 10 minutes than in those baked at the higher temperatures for shorter periods of time. Also the maximum internal temperature reached in meringues baked at 325° F. (163° C.) appeared to be somewhat greater than in the replicates baked at 375° F. (190.5° C.) and notably greater than in those baked at 425° F. (218° C.). These observations suggest that the length of time a meringue is held at a temperature within the coagulation range of the meringue mixture is a more important factor in minimizing leakage than the maximum internal temperature reached. The dominant effect of the nature of the base in determining the degree of coagulation of a meringue is emphasized by the fact that less leakage was obtained with the highest baking temperature and a hot base than with a cold base or paper and a baking temperature of either 325° or 375° F. (163° or 190.5° C.).

No wholly satisfactory explanation for the observed effect of quality of eggs or the addition of potassium acid tartrate on the extent of leakage can be given. As far as is known coagulation temperature of egg white protein is not appreciably affected by egg quality. Coagulation is influenced by pH, and so the addition of potassium acid tartrate might be expected to lower the coagulation temperature of the meringue and decrease the extent of leakage. Such a decrease was apparent, especially in meringues from the lower grades of eggs where the greatest decrease in pH was observed owing to the addition of potassium acid tartrate. Considering the entire experiment, however, no significant correlation was established between pH and the extent of leakage.

Another possible explanation for leakage is that, owing to instability of the foam, liquid separates from the meringue. The differences observed in the extent of leakage in meringues made from eggs of different qualities can not be explained on this basis since no appreciable difference was seen in foam stability as judged by the amount of drainage in one hour. The addition of potassium acid tartrate, however, did influence foam stability.

A class problem was conducted to test the findings of this study. Meringuges, made as nearly alike as was possible by 135 students, were baked on cream pie fillings of varying temperatures. The fillings were chocolate, butterscotch, lemon, cocoanut and banana. The fillings were at the following ranges of temperature: 140° to 176° F., 104° to 131° F., 59° to 77° F. The three baking temperatures of this study were repeated.

The class results on leakage were not consistent with those of the controlled experiment in which a starch-thickened water mixture was used as the base. There appeared to be greater variation owing to the kind of filling than to either the temperature of the filling or to the baking time and temperature. This indicates that the filling as well as the meringue is a factor in determining leakage. It may be influenced by the filling ingredients and their proportions or a rupturing of the starch granules during the cooking process. Further research is required before any explanation can be given for this possible source of leakage in meringue pies.

Beading. The observations of this study suggest that beading of meringues is caused by over-coagulation of the protein with a subsequent loss of some of the adsorbed liquid which appears as beads on the surface. The most pronounced beading was observed on meringues baked on a hot base. A high degree of coagulation would be expected with these meringues since heat was applied from both top and bottom surfaces and the rate of cooling was relatively slow. One can account for the small amount of beading noted on the other meringues by the slower rate of heating and rapid cooling on a cold base, and the rapid cooling on paper. Both would result in a lesser degree of coagulation.

A second factor which appeared to influence the extent of beading was the addition of potassium acid tartrate. Except for the Fancy Grade eggs the extent of beading was increased with each increment of potassium acid tartrate; the effect was most marked with the Grade C eggs. It will be recalled that these were also the samples which showed the greatest lowering of pH. This tempts one to offer as explanation for the increase in beading, the acceleration of the rate of coagulation of egg protein with the addition of acid. It is admitted that the failure to obtain a correlation between the extent of beading and pH somewhat discredits this reasoning.

In regard to the effect of baking temperature and time it has been pointed out that the maximum internal temperature was greater in meringues baked at 325° F. (163° C.) for 10 minutes than in those baked at 425° F. (218° C.) for 4½ minutes. This might be expected to cause a greater degree of coagulation in the meringues baked at 325° F. (163° C.). Actually a tendency was observed for somewhat greater beading at this temperature. The fact that meringues baked at 375° F. (190.5° C.) for 7 minutes showed the greatest beading

is not consistent with this explanation and indicates the need for further study.

In regard to beading the results of the class experiment involving 135 students were in excellent agreement with the results of the controlled study. Regardless of the type of filling numerous beads were observed on the meringues baked on hot fillings (140° to 176° F.) and relatively few on those baked on warm (104° to 131° F.) or cold (59° to 77° F.) fillings. Also fewer beads were observed on meringues baked at 425° F. (218° C.) than on those baked at 325° F. (163° C.).

Summary

The effect of quality of eggs, of addition of potassium acid tartrate, of type and temperature of the base upon which a meringue is baked, and of baking time and temperature upon the quality of soft meringues was studied. Main emphasis was given to possible causes of beading and leakage of the baked product.

The results suggest that a desirable degree of coagulation is an important factor in controlling the amount of leakage and beading in a soft meringue. Leakage was excessive under conditions which would tend to result in incomplete coagulation; beading was excessive under conditions which would favor over-coagulation.

The dominant factor in determining the extent of leakage and beading was the temperature of the base on which the meringue was baked. Greatest leakage and least beading were observed with meringues baked on fillings having a temperature of about 59° F. (15° C.). Evidence indicates that the internal temperature reached in these meringues was not above 158° F. (70° C.); they cooled relatively rapidly. Greatest beading and least leakage were observed with fillings having a temperature of 131° to 140° F. (55° to 60° C.). In these the internal temperature was at least 18° F. (10° C.) higher than in the replicates baked on bases at 59° F. (15° C.); they cooled relatively slowly. Checked under class room conditions similar results on beading were obtained but the type of filling appeared to be even more important than its temperature in influencing leakage, suggesting the need for a controlled experiment with different types of fillings.

On fillings at 59° F. (15° C.), where beading was at a minimum, less leakage occurred in meringues baked at 325° F. (163° C.) for 10 minutes than at 425° F. (218° C.) for four and one-half minutes. On fillings at 131° to 140° F. (55° to 60° C.) where leakage was at a minimum, less beading occurred at 425° F. than at 325° F. Regardless of filling temperature, meringues baked at 425° F. were always more tender and showed less stickiness when cut than those baked at the lower temperatures. The maximum internal temperature of meringues baked at 425° F. was notably less than at 325° F. That the results obtained using a baking temperature of 375° (190.5° C.) for seven minutes were not intermediate between those obtained at 325° and 425° F. cannot be explained at this time.

The extent to which egg quality and the addition or omission of potassium acid tartrate affected leakage appeared to be of little significance compared with the other factors studied. Grade of eggs used was insignificant in relation to the beading of meringues; but the addition of potassium acid tartrate, especially to eggs of lower grades, tended to increase the extent of beading.

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An Improved Heated-Thermocouple Anemometer for Use in Air-Blast Freezers[a]

E. LOWE AND J. R. HAWES

Western Regional Research Laboratory,[b] Albany, California

An improved heated-thermocouple anemometer of the type originally devised by W. V. Hukill was developed for use in measuring the air velocity in air-blast freezers. A method was devised for adjusting the calibration of each individual anemometer to fit a common calibration curve, making possible the use of any number of anemometers interchangeably.

It is generally recognized that air velocity and air distribution have a considerable effect upon the freezing rate of the product in air-blast freezers. Recent tests of a commercial freezer show that the time required to to reduce the center temperature of lima beans in 12-oz. packages from ambient temperature to 0° F. varied from 4⅓ to 8¾ hours. One of the causes of this undesirable wide variation in freezing time was the poor air velocity distribution in the freezer, which ranged from approximately 600 feet per minute to less than 100 feet per minute over the test packages.

In order to develop a better understanding of the effect of air velocity and other variables upon the time necessary to freeze food products, freezing-rate studies are being conducted at this Laboratory. In connection with these studies an instrument was required for accurately measuring the velocity of the air moving past the product in the freezer. The need was for an anemometer with the following characteristics:

(1) The instrument must accurately indicate air velocities over the range normally encountered in air-blast freezers, namely, from 10 to 2500 feet per minute.

(2) The sensing element of the instrument must be small enough so that it can be located in the space between the trays in the freezer, as well as between the various test packages on the tray.

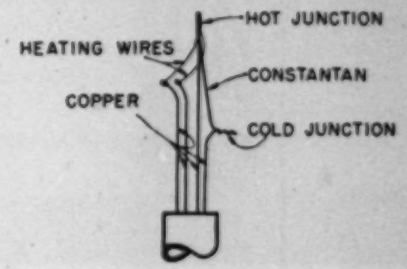
(3) The instrument must be of the remote reading type, since measurements must be made in restricted or confined quarters where the presence of an observer is not feasible or would affect the accuracy of the reading. The remote reading feature is desirable also because the measurements will be made over a long period of time at air temperatures ranging from 0° F. to —40° F.

(4) The accuracy of the instrument must be unaffected by age or sudden changes in temperature.

(5) The velocity response of the instrument must be substantially independent of the direction of air approach.

(6) The design of the instrument must be such that it will be convenient to employ a multiplicity of sensing elements in the freezer at the same time.

Work was undertaken to develop an anemometer with these characteristics when it became apparent that because of one limitation or another, the usual instruments for measuring air velocity would not be satisfactory. The instrument developed is a modified and improved version of the heated-thermocouple anemometer originally devised by W. V. Hukill of the Bureau of Plant Industry, Soils, and Agricultural Engineering, U. S. Department of Agriculture *(1)*. The sensing element of the instrument is shown in Figure 1, and consists essentially of two thermocouple junctions in



"DETAILS OF HEATED-THERMOCOUPLE ANEMOMETER"

FIG. 1.

opposition, with one of the junctions heated externally by a coil of heating wire wound around the junction. The difference in temperature between the two junctions is a function of the rate of cooling of the heated junction as affected by the velocity of the air moving past the junction. This difference in temperature causes a difference in electromotive force between the couples, which is detected and measured by a standard potentiometer circuit. By applying different voltages to the external heating wire it is possible to measure air velocities over a wide range.

Figure 2 is a photograph of the sensing element. The two thermocouple junctions are made from 22-gage enameled copper and constantan thermocouple wires twisted together and soldered. The heated junction is then dipped in Bakelite[c] varnish, dried and wound with 30 turns of No. 40 enameled contstantan wire. Physically, the element is approximately 3″ long by ⅞″ wide by ¼″ thick. For convenience in handling and positioning of the anemometer in the freezer, the sensing element is mounted on a 6-prong miniature plug. The thermocouple and heater wires each occupy two of the connections, while the last two connections accommodate a standard thermocouple junction for measuring the temperature of the air in the freezer.

The miniature plugs simplify the problem of installing a large number of sensing elements in a test freezer. This is especially true where long connecting leads are

[a] Presented at the Annual Meeting of the Northern California Section of the Institute of Food Technologists, December 2, 1948, Albany, California.

[b] Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, U. S. Department of Agriculture.

[c] Mention of this product does not imply that it is endorsed or recommended by the Department of Agriculture over others of a similar nature not mentioned.

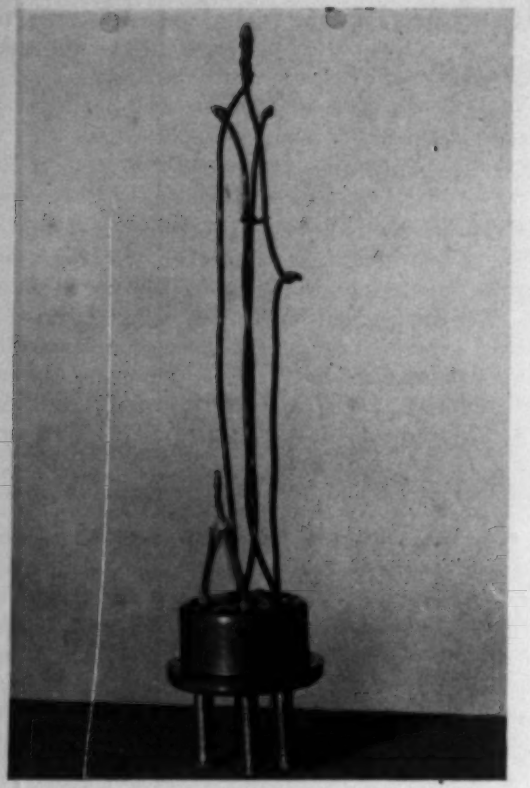


FIG. 2.

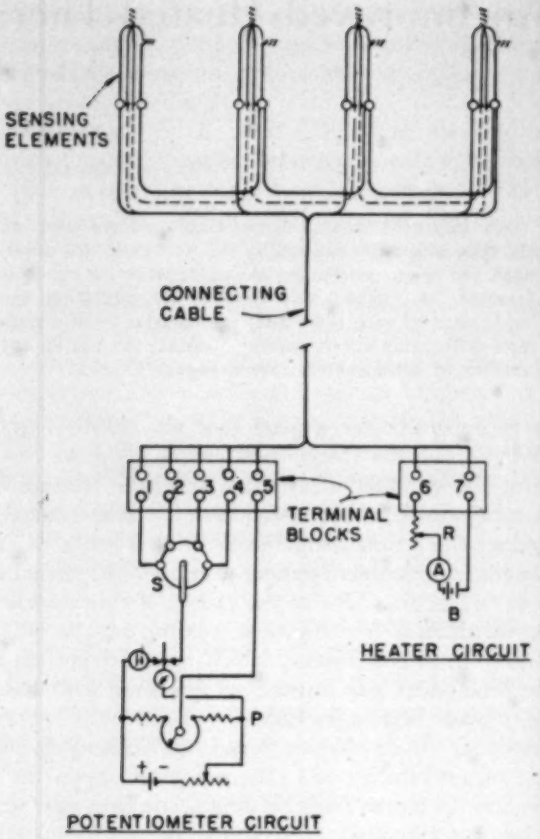


FIG. 3. Typical circuit diagram for a group of 4 heated-thermocouple anemometers.

required. The wiring and plug receptacles can be installed in the freezer prior to the start of the test run and the elements plugged in later as they are needed. This procedure minimizes the possibility of damaging the sensing elements while the instrument is being installed or during periods when the instrument is not in use. Obviously, the use of plugs also facilitates replacement of defective sensing elements.

Figure 3 shows a typical circuit diagram for a multiple-point installation. The velocity at any one of four points in the freezer can be read remotely on potentiometer, P, the particular point being determined by the position of selector switch, S. Current for heating the hot junctions is supplied by battery, B, the actual current flowing through the wires being adjusted by variable resistor, R, and indicated on milliammeter, A. The connecting cable between the sensing elements in the freezer and the potentiometer outside can be made as long as desired, and in one actual installation was over fifty feet long. The indicating potentiometer and selector switch shown can be replaced by a multi-point recording potentiometer, in which case the air velocities would be recorded continuously.

Figure 4 shows typical calibration curves for two heated-thermocouple anemometers. The two curves show that the calibrations are not identical, simply because it is virtually impossible to construct two or more sensing elements exactly alike. This would indicate that specific calibration curves would be necessary for each individual element. Experimentally, however, it was found that by trimming the length of the heated junction of sensing element No. 2 so as to decrease the area exposed to heat transfer, it is possible to increase

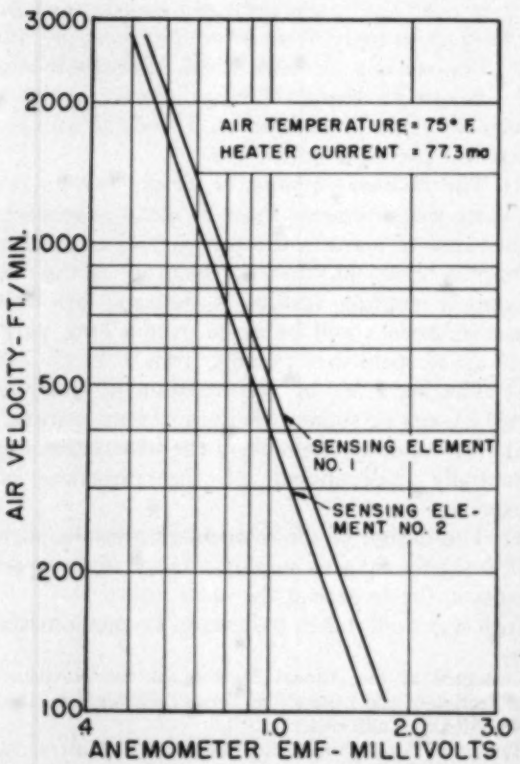


FIG. 4. EMF vs. air velocity.

the millivolt or temperature difference produced by that particular element so that its calibration coincides with that of sensing element No. 1. In the same manner all similar anemometers can be fitted to one calibration curve for any given air temperature, making possible the use of any number of anemometers interchangeably.

Figure 5 shows the calibration curve for air at —10, +75, +120, and +170° F. The e.m.f.-air velocity

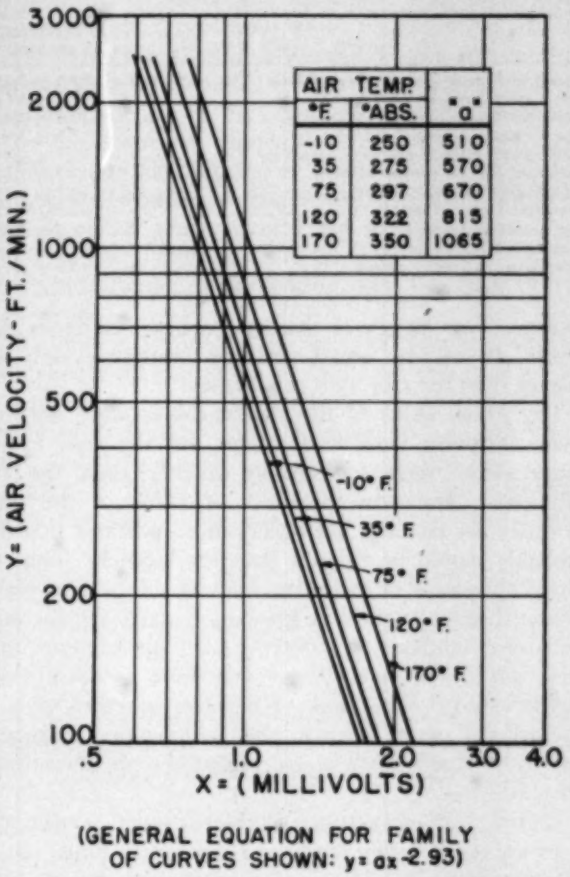


FIG. 5. EMF vs. air velocity at various air temperatures.

relationship for the particular family of curves shown can be expressed by the equation $y = ax^{-2.93}$, where y = air velocity in feet per minute, x = millivolt reading, and the constant a has the values shown on the figure. Graphically, the relationship between air temperature and constant a is shown in Figure 6. No simple mathematical relationship exists between the two. Hukill *(2)* has suggested that individual calibration curves at the various air temperatures might be

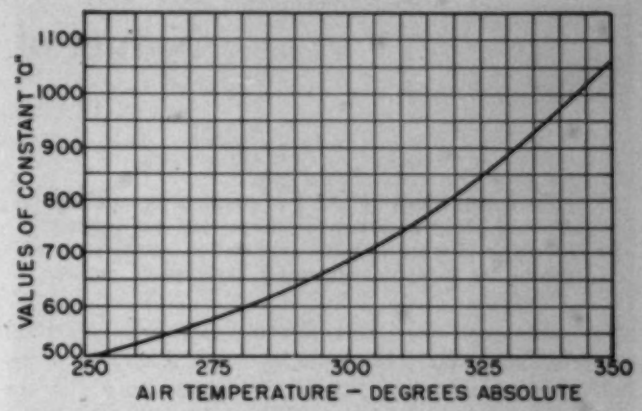


FIG. 6. Air temperature vs. constant "a" in the equation $y = ax^{-2.93}$.

made to coincide by adjusting the heating current in accordance with the existing air temperature. If this is possible, and indications are that it should be, then all air temperature measurements made by any particular set of heated-thermocouple anemometers could be expressed by a simple exponential equation of the form $y = ax^n$.

Although designed primarily to fill a basic need in connection with research studies of the freezing rates of fruits and vegetables, the anemometer described may find further application in commercial food processing plants where air is used as a heat transfer medium, for example, freezing and dehydration plants. In all such cases, air velocity and distribution play an important role. The instrument described offers a convenient and accurate means for determining the actual air velocities prevailing in such process equipment as air-blast freezers and driers. The anemometer has been used for just such a purpose. During recent tests of a commercial air-blast freezer, fifteen of the sensing elements were used simultaneously to record the local velocities in the freezer. As previously mentioned, the data obtained showed a surprisingly wide variation in air velocities, a factor which no doubt contributed to the comparatively slow and non-uniform rate of freezing which was observed during the tests.

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Observations on the Toughness and Microscopic Nature of Prune Skins[a]

LEONORA HOHL STROHMAIER

Division of Food Technology, University of California, Berkeley, 4, California

The author shows by means of photomicrographs the effect of different processing steps, and particularly the effects of lye, steam and dehydration, on the structure of the skin of four major varieties of prunes.

A year ago at our annual meeting there was presented a preliminary report on some histological investigations which were just being initiated *(2)*. The ultimate object of this line of research is to determine what practical use can be made of a knowledge and understanding of the structural and chemical microscopic changes which food substances undergo during processing. There are many processing problems to which this line of attack may be applicable, but this discussion is limited to prune skin problems.

In recent years the sounder, more sanitary dehydrated prune has practically replaced its earlier sun dried counterpart. Along with the well-known advantages of the new form of this product, there simultaneously arose a new problem—namely, that the skin of the dehydrated fruit is often unpleasantly tough and leathery no matter how the fruit is cooked.

Some of the factors which may influence the texture of prune skins are the actual thickness of the skins, size, structure, and the arrangement of the cells making up the skins, as well as the chemical nature of the cell wall constituents and how these are affected by processing.

The first task was to determine the characteristics of a fresh unprocessed prune skin and to learn something of its nature. Skin samples of four major varieties—French, Imperial, Sugar, and Robe de Sargent—were studied in the fresh state, but most emphasis was placed upon the French and Imperial.

The relative toughness of the fresh skins of these four varieties was found to be as follows:

Imperial	1.10 ± .22
Sugar	1.07 ± .23
Robe de Sargent	1.16 ± .22
French	1.84 ± .26

The figures represent the relative load required to penetrate or puncture the skin with a mechanical penetrometer *(1)*. They represent arithmetic means of 100 readings. They do not represent any given weight unit, but are on an arbitrary scale, which remains comparable as long as the instrument is set up with a constant load traveling along a graduated beam. It is interesting to note the significant difference between the French prune and the remaining three varieties; French prune skins are markedly tougher than the others.

Table 1 shows the results of micrometer measurements of thickness of French and Imperial prunes collected from two different ranches in the Sacramento Valley. Two stages of maturity are shown from each ranch. It will be noted that the immature skin was thicker than the ripe skin in all cases.

TABLE 1

Thickness of Skins of Fresh Sacramento Valley French and Imperial Prunes from Three Ranches

	(Thousandths of an inch)
Immature French prunes—Live Oak	13.1 ± 2.32
Ripe French prunes—Live Oak	10.4 ± 3.14
Immature French prunes—Yuba City	13.8 ± 3.03
Ripe French prunes—Yuba City	7.2 ± 3.90
Orchard run French prunes—Yuba City	11.7 ± 2.84
Immature French prunes—Colusa	11.2 ± 3.13
Ripe French prunes—Colusa	10.7 ± 3.23
Immature Imperial prunes—Live Oak	12.2 ± 3.82
Ripe Imperial prunes—Live Oak	6.0 ± 2.26
Immature Imperial prunes—Yuba City	9.9 ± 3.09
Ripe Imperial prunes—Yuba City	5.8 ± 2.45

The green skins of the two varieties were not very much different from each other, but the ripe French prune skins were measurably thicker than the ripe Imperials. Data on thickness of skins of the other varieties are lacking, but observation indicates that they probably would be thinner than the French. Thus the actual thickness of the skins does not appear to play a role in their texture. The growing district and the other cultural conditions undoubtedly have an influence upon this factor of skin thickness but these points all need further study. The time of harvest, whether early or late in the prune season, also influences the relative toughness and thickness, as well as the pigmentation of the skin.

Figure 1 shows a thin (15 microns) cross section of a prune skin when prepared for sectioning by the paraffin method as described in standard texts on microtechnique such as Johansen *(3)* and stained by some

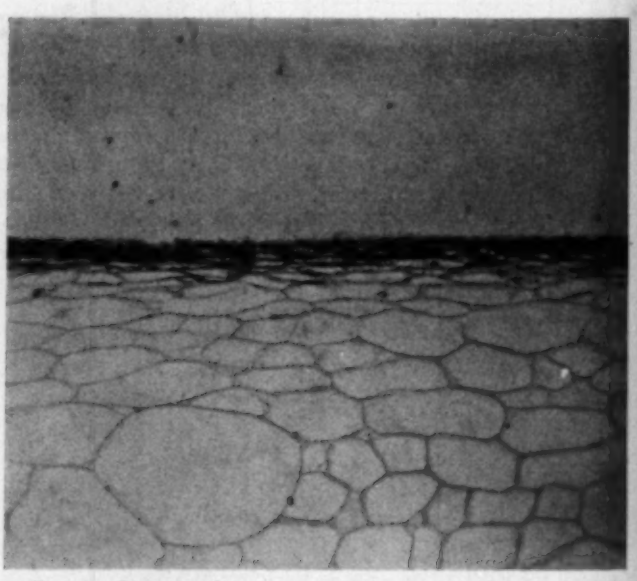

FIG. 1. Cross section of fresh untreated French prune skin with some of underlying flesh parenchyma still attached. X107.

[a] Paper presented at the Annual Meeting Northern California Section, of the Institute of Food Technologists, December 2, 1948.

suitable cell wall stain such as Delafield's hematoxylin. In preparing this material for sectioning the skin was simply cut off with a razor allowing some of the flesh immediately beneath the skin to remain. When prepared in this way it's difficult to decide where the skin ends and the flesh begins. Most of the other photomicrographs and skin samples for penetrometer and thickness measurements were prepared by removing the skin as it peeled off naturally, and then gently scraping off the bits of flesh which adhered to it. In such preparations the skin usually ends in the region about four cell layers below the smallest cells which lie at the outermost surface of the skin. In normal unprocessed prune skin shown in Figure 1, there is seen a thin layer of cuticle and about three or four layers of small cells, the uppermost layer or epidermis being made up of the smallest cells. Then below these three or four layers is this rather indefinite layer of larger, thinner walled cells which grades imperceptibly into the flesh portion of the fruit.

The major portion of the research to date has been concerned with the effect of different processing steps or procedures upon the structure of this skin. The effects of lye, steam and dehydration are the major points shown by the series of photomicrographs.

Figure 2 indicates how very mild lye treatment affects the skin. This was taken from a fresh French

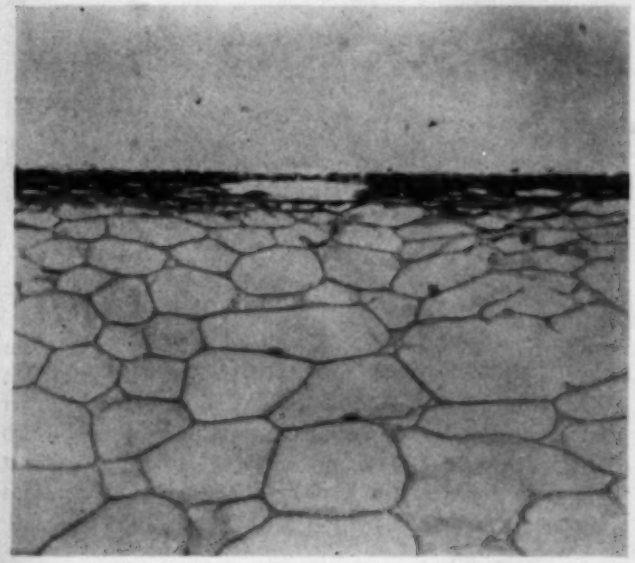

FIG. 2. Cross section of French prune skin after mild lye treatment [0.5 percent NaOH, 30 sec., 150° F. (65.5° C.)]. Some flesh parenchyma still attached. X107.

prune dipped in 0.5 percent NaOH solution at 150° F. (65.5° C.) for thirty seconds. The only change which may be apparent is a few very small breaks in the cuticle layer on the surface.

Figure 3 shows what more drastic lye treatment will do causing large eruptions to be formed in the skin. The breaks seem to occur most commonly in about the second cell layer from the surface. This is characteristic of 0.5 percent lye treatment for thirty seconds at 190° F. (88° C.) or above. Increases in lye concentration or time of dip also have a similar effect. If fresh prunes

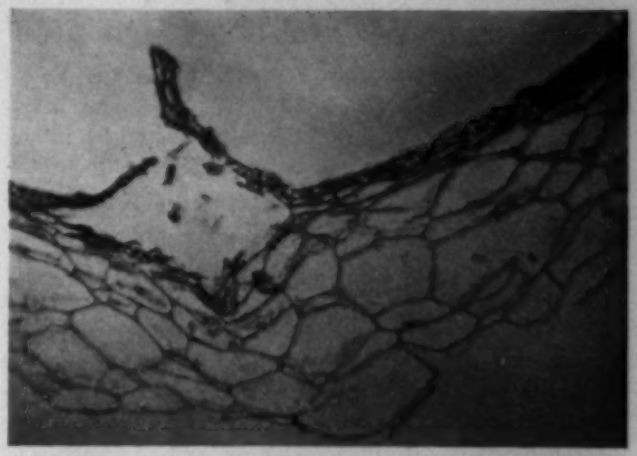

FIG. 3. Cross section of French prune skin after severe lye treatment [0.5 percent NaOH, 30 sec., 190° F. (88° C.)]. X107.

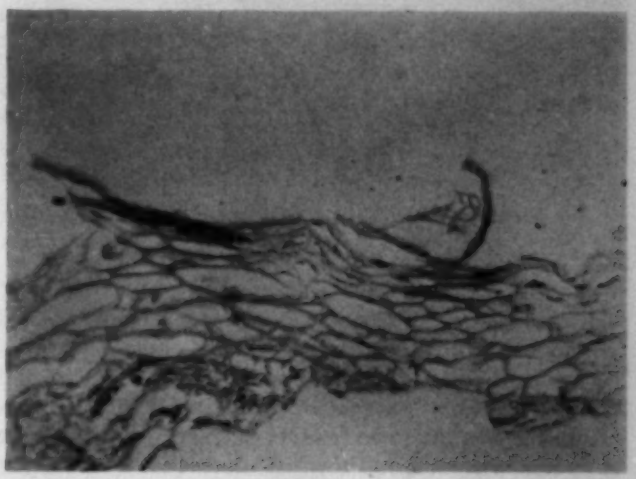

FIG. 4. Cross section of French prune skin after severe lye treatment followed by dehydration at 165° F. (74° C.). X107.

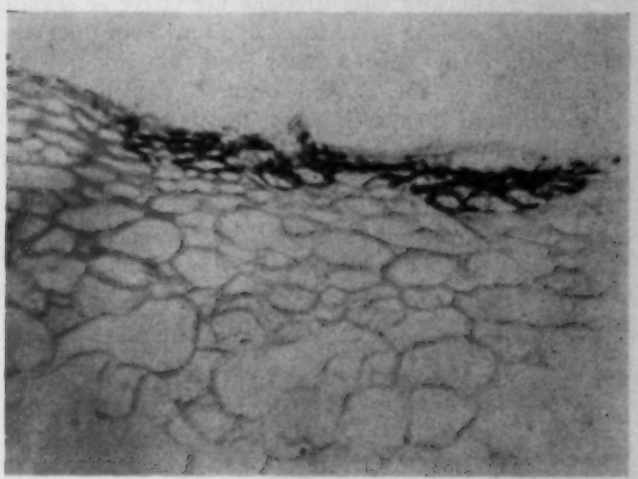

FIG. 5. Cross section of fresh French prune skin, steamed 4 min. X107.

are steamed for four or five minutes, the cross sections of the skins have much the same appearance.

Figure 4 shows a prune skin which has been lye dipped much like the previous one, but has also been dehydrated. The severed epidermal layer has curled away from the underlying tissue.

Figure 5 shows the effects of steam upon fresh French prune skins.

Figure 6 shows how the skin is affected when an otherwise untreated prune skin has been half dehydrated. Note again how the epidermal layer is beginning to break away from the cell layer immediately below it, but there have been no actual breaks in the epidermal layer itself up to this point.

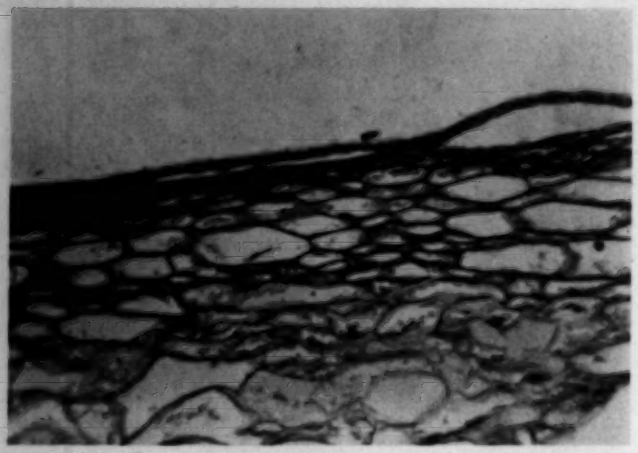

FIG. 6. Cross section of half dehydrated French prune skin. X107.

In Figure 7 the effect of steaming a similar half dehydrated prune may be observed. The loosened uppermost layers tend to break off.

Figure 8 shows the skin of a prune which was half dehydrated, then steamed, and finally dehydration was completed to a little over twenty percent moisture. That skin was wrinkled and torn so much that it could not be made to lie flat when being sectioned and prepared for study.

Table 2 indicates data to show the effect of several different processing procedures upon the skin thickness and texture by tenderometer and by organoleptic examination of the cooked prunes. Six processing methods were compared. For the penetrometer and thickness measurements the skin was removed and carefully scraped. The table shows that the sample which was dehydrated to twenty percent moisture without any other treatment was markedly tougher than any

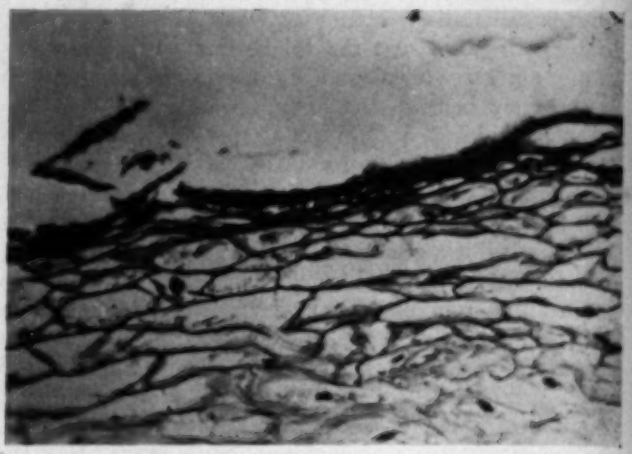

FIG. 7. Cross section of half dehydrated and steamed French prune skin. X107.

FIG. 8. Cross section of French prune skin which was half dehydrated, steamed for 4 min., and then completely dehydrated.

of the samples which had received steam, hot water, or lye treatments.

Figure 9 shows the skin of a French prune which has been dehydrated at 165° F. (74° C.) without further treatment. The cells of such a prune are mostly intact and compactly arranged in contrast with those which have receieved some steam or lye treatment. This fact is also reflected in the greater tenderness of the skins

TABLE 2

Effect of Processing Upon Some Properties of French Prune Skins

Treatment of Sample	Skin thickness	Penetrometer readings	Organoleptic observations on cooked prunes
	(Thousandths of an inch)		
Steamed 4 minutes and dehydrated at 165° F. (74° C.)	5.1 ± 1.5	1.31 ± .85	Tender to all tasters
Half dehydrated at 165° F. (74° C.), steamed 5 minutes and dehydration completed to 20 percent moisture at 150° F. (65.5° C.)	5.4 ± 1.6	1.41 ± .75	No definite trend
Dehydrated to 30 percent moisture. Steamed 10 minutes	5.4 ± 1.2	1.46 ± .77	No definite trend
Dehydrated to 20 percent moisture	6.8 ± 1.4	2.97 ± .74	Toughest skin to majority of tasters
Dipped in water at 212° F. (100° C.). Dehydrated at 165° F. (74° C.)		.87 ± .30	Tender as compared to above treatments. Tough when compared to lye dipped samples
Dipped into 0.5 percent NaOH solution at 212° F. (100° C.) for 30 seconds. Dehydrated at 165° F. (74° C.)		.43 ± .24	Tender and disintegrated

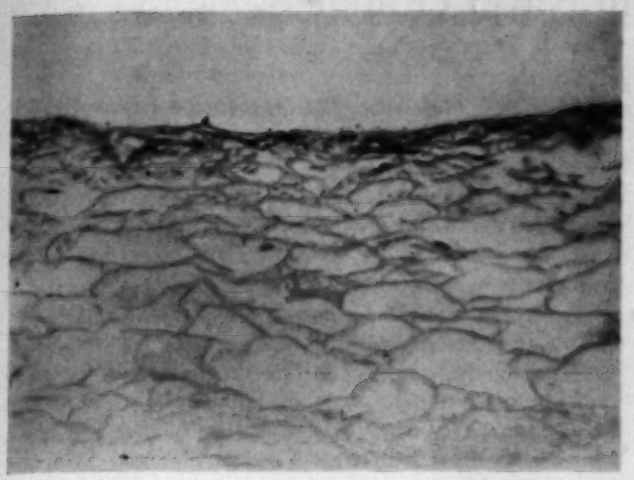

FIG. 9. Cross section of French prune skin which has been dehydrated at 165° F. (74° C.) without further treatment. X107.

receiving these special treatments causing rupture of the cell structure.

Hot water dipping of prunes was also compared with lye dipping and steaming. Microscopically, hot water dipped fruit showed very much milder checking and rupture of the surface cell structure. Organoleptically and by penetrometer tests they also were definitely tougher than lye dipped skins.

The tentative conclusion to date is that dehydrated prune skins, especially French prune skins, will be tough unless they are given some rather drastic treatment such as steaming or lye dipping. Such treatment is more effective in tenderizing prune skins if applied before dehydration is started rather than at the half dry or completely dehydrated stage.

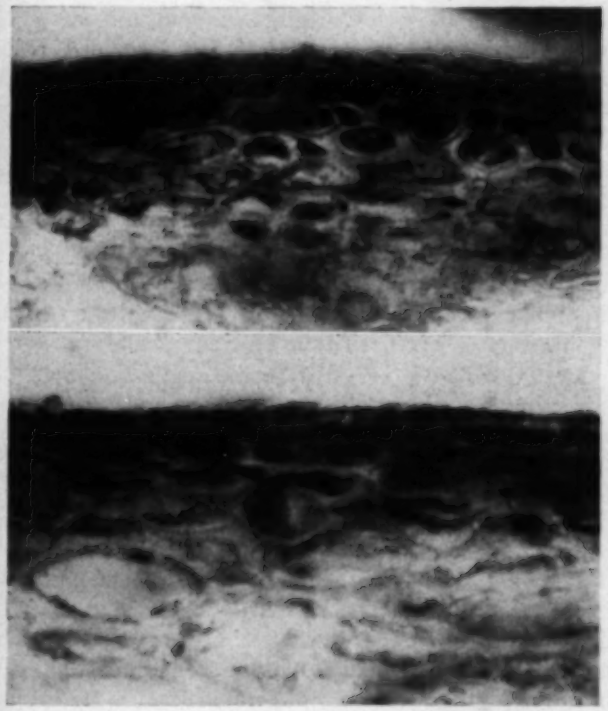

FIG. 10. Free-hand cross sections of immature (above) and mature (below) fresh French prune skins. X approx. 200.

Another property of prune skins which has been of interest to study microscopically has been the pigment and its distribution in the fresh skin as well as its behavior during dehydration. Figure 10 shows free-hand sections of fresh immature and mature French prunes. Distribution of the pigmented cells at the two stages should be noted.

Figure 11 shows a cross section of a mature Imperial prune skin. The pigment is a water soluble red, probably anthocyanin.

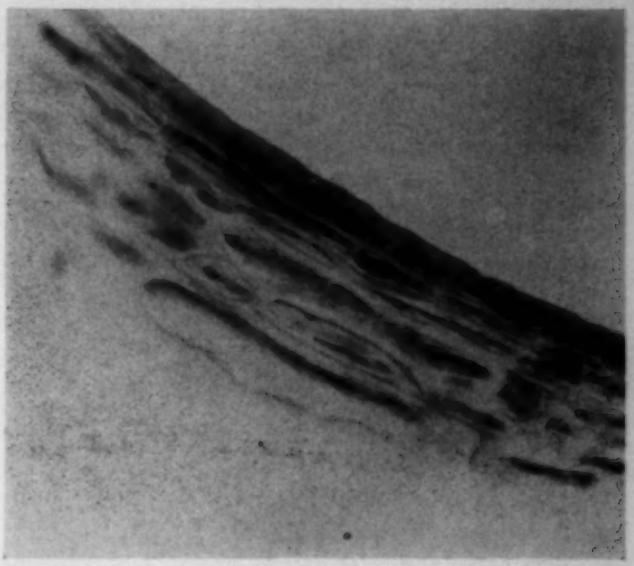

FIG. 11. Free-hand cross section of fresh Imperial prune skin. X approx. 200.

Pigment behavior during dehydration at high, medium, and low temperatures was observed and followed microscopically and macroscopically and it was found that there was less discoloration of the flesh immediately below the skin in prunes dehydrated at 170° F. (77° C.) or above, than in similar prunes dehydrated at 140° F. (60° C.). The discoloration observed in the low temperature lot appeared to be more enzymatic browning than red skin discoloration.

Acknowledgments

The author wishes to acknowledge with sincere thanks the assistance of her husband, Erwin Strohmaier, in making the photomicrographs, and of Dr. Florence Pen Ho, visiting scholar in Food Technology, in preparation of samples. The use of the penetrometer was generously permitted by Miss Mildred Boggs of the Western Regional Research Laboratory, U. S. Department of Agriculture at Albany, California.

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The Bitter Principle in Navel Oranges[a]

OLIVER H. EMERSON

Enzyme Research Division, Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, United States Department of Agriculture, Albany, California

A bitter principle affects the value of Navel oranges for juice products. The chemical and physical properties of this limonin were studied and the effect of natural and stimulated ripening was observed under limited conditions.

A serious problem to the California orange growers is the fact that Navel oranges sometimes give a juice which, in the course of a few hours, becomes unpalatably bitter. This takes place when the juice is canned or frozen or kept in a refrigerator, and has seriously discouraged the processing of this variety of oranges. This bitterness is most marked with early fruit, and usually becomes less noticeable as the season progresses. The first chemical study of the bitter principle occurring in Navel oranges was done by Higby *(4, 5)* who concluded that the substance was limonin and/or isolimonin. Limonin, $C_{26}H_{30}O_8$ (not to be confused with the essential oil limonene, $C_{10}H_{16}$) was discovered by Bernay in 1841, and has since been found in a number of citrus and related rutaceous plants, occurring variously in the fleshy part of the fruit, the seeds, the bark, and the roots *(9)*. It is a beautifully crystalline substance melting at 300° C., soluble to various degrees in organic solvents. Higby considered it to be about as bitter as quinine, but its solubility in water is so slight that only very finely divided material, placed on the tongue, tastes appreciably bitter. The bitterness in Navel juice must be due to a colloidal solution, stabilized only too well by the various juice constituents. Aside from its bitterness, limonin appears inert physiologically.[b]

The chemistry of limonin has recently been discussed by Geissman and Tulagin *(3)*. While our knowledge of this subject is still very fragmentary, the fact of immediate concern to the orange grower is that limonin is a dilactone. Lactones are cyclic esters, and like ordinary esters, they are readily cleaved by alkali, giving salts of hydroxy acids. The dihydroxy-diacid from limonin, to which we shall refer as limonic acid, is water-soluble and not bitter, but is stable only in the form of its salts. On acidification it readily cyclizes, regenerating limonin. This change readily takes place at pH 3.6, the usual pH of orange juice.

Isolimonin is a name given by Koller and Czerny *(7)* to a substance they isolated from orange seeds which they believed isomeric with limonin. Lack of material prevented more than a cursory examination, and while it has been reported by other investigators *(8, 10)*, its characterization leaves much to be desired.

Our first task was to repeat the work of Higby to determine whether the bitter substance of Navel orange juice is limonin, or isolimonin, or a mixture of the two. We obtained as the sole isolable bitter substance, limonin, agreeing with the description of that substance by other workers, and with material obtained from the seeds of Valencia oranges, lemons and grapefruit. Our interest in seeds lay in a desire to obtain some isolimonin. From the seeds of oranges and lemons we obtained a substance resembling somewhat the description of isolimonin, but our product was not an isomer of limonin. It had the empirical formula $C_{28}H_{34}O_9$, so we named it nomilin *(2)*. Nomilin resembles limonin in being a beautifully crystalline, high melting substance with a similar absorption spectrum and, in general, similar solubilities. Like limonin, it possesses a rather unreactive carbonyl group and is a dilactone, but its reaction with alkali is more complex than that of limonin, for acetic acid is split out and there is formed on acidification a lactone acid, $C_{26}H_{32}O_8$. Like limonin, nomilin appears to be inert physiologically under the conditions tried.[b]

Nomilin may be an important constituent of citrus seed bitters. A batch of orange seeds yielded 1.4 g. of limonin and 0.8 g. of nomilin per kilo, while some lemon seeds yielded likewise 0.6 g. of limonin and 0.8 g. of nomilin. A batch of grapefruit seeds yielded more limonin, 2.4 g./kilo, but no nomilin. Nomilin does not appear to occur in significant amounts in the juice of early Navels, or of very immature Valencias.

There are several phytochemical problems concerning limonin which are of interest to the orange grower. What is the non-bitter precursor of limonin in the Navel orange? Since it is the early season fruits which are most troubled with bitterness, is the formation of limonin a feature of immaturity? If so, can the trouble be ameliorated by an artificial ripening of the fruit?

Higby made the very reasonable suggestion that the non-bitter limonin precursor is limonic acid, or perhaps a derivative of it such as a glycoside, which, in the intact fruit, exists in tissues whose pH is considerably higher than the juice and permits it to be stable. By oxidative degradation of limonin, Geissman and Tulagin obtained a lactone acid, $C_{25}H_{28}O_9$, which was not bitter; thus it is possible that one of the pair of lactone acids formed by the opening of only one lactone ring of limonin might be the non-bitter precursor. We have done some work indicating that the formation of limonin in Navel orange juice is more rapid than is its formation from limonic acid at the same pH in water, or in

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[a] Enzyme Research Division Contribution No. 120.

[b] Dr. M. E. Simpson of the Institute of Experimental Biology, University of California, fed 10 mg. of limonin suspended in ethyl laurate daily to a number of rats for a period of ten days. No physiological responses were observed, nor were any changes noted at autopsy. Dr. J. C. Lewis of the Western Regional Research Laboratory likewise found limonin and nomilin to be devoid of action against a number of bacteria. Dr. R. H. Wilson of the Pharmacology Division fed nomilin incorporated into the diet at a level of 1/1000 to a group of young rats for one week without impairment of the growth rate or other untoward effect.

Valencia juice which did not naturally turn bitter. If this observation is correct, unless the process in Navel juice is catalyzed by enzymes in addition to hydrogen ion, the precursor cannot be limonic acid, but might be the more rapidly cyclizing member of the pair of lactone acids, or perhaps be a glycoside. In view of the sensitivity of the precursor to acid, and the possible presence of a lactone group sensitive to alkali, the isolation of the precursor might be a matter of considerable difficulty.

The idea that the appearance of limonin in Navel juice is a feature of immaturity receives considerable support from finding that very immature Valencias also develop limonin.[c] A batch of 25 kilos of Valencias collected at Riverside, California, about the middle of November, 1947, yielded 8 liters of juice from which was obtained 140 mg. of limonin. Still more (2.27 g.) was obtained from the peel. However, in January, 1948, we found that the Valencia juice no longer turned perceptibly bitter. Hence the disappearance of limonin from Navel juice has a parallel in Valencia juice, except that in the case of the latter, the disappearance takes place long before the fruit is ready to eat.

Limonin persists in Valencia peel long after it has disappeared from the juice. In May we found nearly as much limonin in Valencia peel as we had found in November. In order to see if this limonin were preformed, or existed in the form of a water soluble precursor, fresh peel (850 g.) from two dozen oranges was ground in a meat grinder and collected in 4 liters of a 1 percent sodium acetate solution at pH 6.0. At this pH the cyclization of the precursor is practically completely arrested. The ground peel was thoroughly extracted with this solution and then with water. After extraction with butanol to remove any colloidal limonin, the united aqueous extract was strongly acidified (pH 2.0) to cyclize the precursor, and after standing was again thoroughly extracted with butanol. From this was obtained 16 mg. of limonin which may be regarded as representing precursor limonin. The washed meal was then thoroughly extracted with butanol, yielding 159 mg. of limonin which must have been preformed.

It was felt of interest to see if this preformed limonin would remain in the peel or be removed as the fruit further matured. A similar lot of peel gathered from fruit collected a month later yielded only 25 mg. of limonin. How does the plant transport such an insoluble substance as limonin? Is the conversion of the water soluble "precursor" to limonin a reversible process?

With this evidence that the development of limonin in Navel orange juice may be a feature of immaturity, we have tried the effect of artificial ripening of the fruit by treatment with ethylene and acetylene. Acetylene is being quite extensively used in Hawaii to speed up the ripening of pineapples. Ethylene is well known to stimulate the respiration of many fruit, increasing the oxygen uptake and CO_2 output. Also, it will cause changes in the pectin content of the albedo of oranges, and effect a disappearance of chlorophyll from the flavedo, so that the fruit will have a brighter orange color. In fact it is a widely used practice to treat oranges with ethylene before marketing in order to improve their color. The fruit growers have studied the effect of ethylene on oranges under a variety of conditions in the processing plants in the hope of mitigating the tendency to turn bitter. Experiments made in this laboratory showed that relatively heavy doses of ethylene did remove the bitter substances, but unfortunately under the drastic conditions used other undesirable flavors were formed. Accordingly we tried the effect of ethylene and acetylene on fruit still on the tree, in the hope that longer time might effect a more natural, but at the same time accelerated ripening. About the middle of November in 1947, and again in 1948, we treated some trees at the University of California Citrus Experiment Station at Riverside with ethylene and acetylene in concentrations of 1/500 and 1/1000 using a neoprene tent [d]. The treatments varied from one hour to 90 minutes. Some of the trees received one treatment, others daily treatments on three successive days. The field temperatures were normal for the season, being around 40° F. early in the morning, and rising to 70° F. later in the day. In general we avoided the warmest portions of the day, because of fear that the much higher temperatures under the tent might be a disturbing factor. I wish I could report some spectacular results, but the treatment as we carried it out appears to have had little effect; in fact, the best fruit came from a control tree. On account of the individual variation in the trees, which was greater than we had anticipated, the experiment would have to be done on a considerably larger scale in order to give significant results.

In addition, we have been doing some work on the chemical constitution of limonin, about which little enough is known. It is well established that the substance has the empirical formula $C_{26}H_{30}O_8$, is a dilactone and contains no free hydroxyl groups, or methoxyl groups. To ketone reagents it is generally unreactive, but by the technique of Bachmann and Boatner *(1)* using pyridine, we prepared a beautifully crystalline oxime showing that the substance contains a carbonyl group, a fact suggested by its absorption spectrum. The remaining three oxygen atoms are very probably cyclic ethers, and Geissman and Tulagin's degradation of limonin by means of hydriodic acid to a substance having the same number of carbon atoms, but with two less oxygen atoms, shows that at least two of these three oxygen atoms must be cyclic ethers.

An important question concerning the structure of a substance is the number of carbocyclic rings and double bonds. A saturated hydrocarbon structure without rings has the general formula C_nH_{2n+2}, and for each ring or double bond two hydrogens are subtracted. Hence it follows from its empirical formula that limonin must

[c] Dr. J. F. Kefford of the Council for Scientific and Industrial Research, Homebush, N. S. W., Australia, told us of finding limonin in very immature Australian Valencias.

[d] We wish to express our sincerest thanks to Dean C. B. Hutchinson and Professor L. D. Batchelor of the University of California, and their associates, for their most kind cooperation without which this work would not have been possible.